

An illustration of a desert landscape. In the background, there are three palm trees of varying heights against a light blue sky. The ground is a mix of light and dark brown patches, representing sand and soil. In the foreground, a large, detailed red imported fire ant is shown from a top-down perspective, facing left. The ant has a reddish-brown body, six legs, and two antennae. The text of the conference title is overlaid on the upper right portion of the illustration.

# Annual Red Imported Fire Ant Conference

Palm Springs, CA

March 30 -- April 1, 2003

Hosted by UC Riverside's  
College of Natural & Agricultural Science  
and  
The Coachella Valley Mosquito and Vector Control District



# *Proceedings of the*

## **Red Imported Fire ant Conference**

March 30 – April 1, 2003

Palm Springs, California

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Department of Entomology  
UC Riverside

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## Acknowledgments

The success of the 2003 Red Imported Fire Ant Conference was mainly a result of the efforts of Les Greenberg for his leadership and overall planning of the conference. Thanks are also due to Carol Lerner for coordinating the conference and proceedings and Jessica Darin for administrative support; UCR's Printing and Reprographics Department for producing this publication; and to Chissa and Suzette from the Palm Springs Riviera Resort & Racquet Club for their assistance and patience with the catering and room accommodations.

Special thanks to John Kabashima for his oversight and assistance, to Joseph Morse for providing administrative assistance (by way of Darren Anderson) in order to maintain the program on the website, and to the Orange County Vector Control District for their donations. Special thanks go to Don Goms, General Manager of the Coachella Valley Mosquito and Vector Control District, for the on-site support of his organization, and to Robert Mann for the design of the meeting logo and t-shirts.

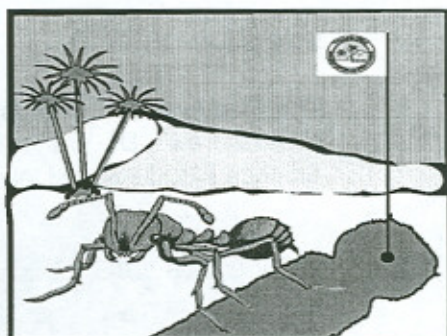
This conference was jointly sponsored by:

- *University of California, Riverside*
- *Coachella Valley Mosquito and Vector Control District*
- *Orange County Vector Control District*
- *UC Center for Invasive Species Research*
- *UC Division of Agricultural and Natural Resources*
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- *Los Angeles County Agricultural Commissioner's Office*



# Annual Red Imported Fire Ant Conference

Palm Springs, CA  
March 30 - April 1, 2003



## Palm Springs Riviera Resort & Racquet Club

### Room Information

Registration at Ballroom Foyer

Posters in VIP Room; Exhibits in Ballroom Foyer

An A/V practice room will be available throughout the conference in Mesquite C

Continental breakfast and refreshments at Conference Center Breezeway

**All talks will be held at the Ballroom Center**

### PROGRAM

#### Sunday, March 30

3:00	10:00	Exhibit and Poster Set-up (VIP Room)
5:00	7:00	Early Registration
6:30	8:00	Welcome Reception (poolside)
12:00	5:00	USDA Organizational Meeting (Oleander I Room)

#### Monday, March 31

7:00	8:00	<b>Continental Breakfast</b>
7:00	12:00	<b>Registration</b>
8:00	9:00	<b>Welcome</b>

Dr. Les Greenberg, Program Chair, Department of Entomology,  
University of California, Riverside

Dr. Joseph Morse, Department of Entomology, University of  
California, Riverside, UC Division of Agricultural and Natural  
Resources Program Leader for Agricultural Policy & Pest  
Management

Donald Goms, General Manager, Coachella Valley Mosquito and  
Vector Control District



8:15

**Guest Presentation**

**The Fire Ant, *Solenopsis invicta*: Is it Really the Beast of Our Nightmares?** Dr. Walter R. Tschinkel. Department of Biological Science Florida State University

**Biology, Ecology, and Detection** – Moderator: Les Greenberg

- 9:00 9:12 **Temporal Foraging Activity of Imported Fire Ants and Native Ants in Northern Mississippi** James T. Vogt, J.T. Reed, and R.L. Brown. USDA, ARS BCPRU
- 9:12 9:24 **Co-occurrence of Ant Species With the Red Imported Fire Ant in South Carolina.** Tim Davis. Clemson University Extension, Craig Allen and Mac Horton. Clemson University
- 9:24 9:36 **Ecological Impacts of Red Imported Fire Ants on the Fauna of Eastern Australia.** Cas Vanderwoude. Australia Fire Ant Control Centre
- 9:36 9:48 **Landscape Maintenance Practices Influence Fire Ant Establishment.** James A. Reinert, M.C. Engelke, J.C. Read, and W.A. Mackay. Department of Entomology, Texas A&M University
- 9:48 10:00 **Observations on Fire Ants and Iridomyrmex Species During an Eradication Program in Brisbane, Australia.** Jo-anne Holley and Kristine Plowman. Fire Ant Control Centre, Department of Primary Industries, Australia
- 10:00 10:12 **Seasonal Effects on Detection of Imported Fire Ant Mounds with Airborne Digital Imagery.** James T. Vogt. USDA, ARS BCPRU
- 10:12 11:00 **BREAK** Posters and Exhibits
- 11:00 11:12 **A Look at the Accomplishments and Ongoing Research of Select Aspects of the Texas RIFA Program.** S.B. Vinson and Colleagues. Texas A&M University, College Station, TX
- 11:12 11:24 **Identification of Gut Bacteria From 4th Instar Red Imported Fire Ant Larvae.** John Peloquin and Les Greenberg. Department of Entomology, University of California, Riverside

**Behavior** – Moderator: Les Greenberg

- 11:24 11:36 **Colony Founding by Multiple Newly Mated Queens of Different Social Forms.** Robert K. Vander Meer and Catherine A Preston. USDA-ARS
- 11:36 11:48 **Fire Ant (*Solenopsis invicta*) Acoustic Stridulation in Response to Phorid Fly (*Psuedacteon tricuspis*) Attack.** James Anderson. University of Mississippi Field Station



11:48 12:00 **New Targets for Disruption of Imported Fire Ant Pheromone Reception.** Robert Renthal, Kalyani Guntur, Daniel Velasquez, and J. Aaron Cassill. University of Texas at San Antonio

12:00 1:00 **HOSTED LUNCH** (Ballroom East)

1:00 2:00 **Posters and Exhibits**

**Biocontrol** – Moderator: John Kabashima

2:00 2:12 **Phorid Fly Release in Arkansas: A County Agent's Perspective.** Jerry Clemons, Mike McCarter, Doug Petty, John Gavin, Kelly Loftin, Donna Shanklin, and John Hopkins. University of Arkansas, Cooperative Extension Service

2:12 2:24 **Prevalence of the Fire Ant Pathogen *Thelohania solenopsae* in Monogyne and Polygyne Colonies.** David Oi, Steven Valles, and Roberto Pereira. USDA-ARS

2:24 2:36 **Phorid Flies in Alabama: A Tale of Two Species.** L. C. 'Fudd' Graham. Department of Entomology and Plant Pathology Auburn University Auburn, AL; Sanford D. Porter. USDA-ARS, CMAVE Gainesville, FL; Vicky E. Bertagnolli. Department of Entomology and Plant Pathology Auburn University Auburn, AL

**Chemical Control** – Moderator: John Kabashima

2:36 2:48 **Evaluation of Soil Residues and Efficacy of Selected Pyrethrin and Pyrethroid Insecticides on Red Imported Fire Ant Mounds.** Nathan Riggs. Texas Cooperative Extension, San Antonio; Wizzie Brown. Texas Cooperative Extension, Austin; Dr. Bart Drees. Texas Cooperative Extension, College Station, TX

2:48 3:00 **Fire Ant Mound and Foraging Suppression by Indoxacarb Bait.** Charles L. Barr. Texas Cooperative Extension, College Station, TX

3:00 3:12 **BREAK**

3:12 3:24 **Spring & Fall Applications of Granular Indoxacarb Baits for Fire Ant Control in Florida.** Richard S. Patterson. University of Florida (*Copy of presentation included on attached CD*)

3:24 3:36 **Repellency and Toxicity of Mint Oil Granules to Red Imported Fire Ants.** Arthur G. Appel and Marla J. Tanley. Department of Entomology and Plant Pathology, Auburn University

**Quarantine and Eradication** – Moderator: John Kabashima

3:36 3:48 **USDA-APHIS/ARS Efforts to Prevent Imported Fire Ant Infestations of Commercial Honey Bee Operations.** R.D. Weeks. USDA-AHPIS; G. Hoffman-DeGrandi. USDA-ARS. (*Abstract not submitted.*)

3:48 4:00 **Update on RIFA Quarantine in CA.** Mohammad Azhar. CDFA



4:00	4:24	<b>Survey, Treatment &amp; Outreach. Eradication Strategy in Orange County.</b> Richard Bowen, Charlie Cassidy, Shana Lowe, and Michael Hearst. Orange County Fire Ant Authority, CA
4:24	5:00	<b>Posters and Exhibits</b>
5:00		<b>ADJOURN FOR DAY – Dinner on your own</b>

## **Tuesday, April 1**

7:00	8:00	<b>Continental Breakfast</b>
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### **USDA Areawide Suppression Project** - Moderator: Roberto Pereira

8:00	8:12	<b>An Integrated Pest Management Approach for the Imported Fire Ant.</b> David F. Williams, USDA-ARS
8:12	8:48	<b>Areawide Fire Ant Control: Project Update.</b> Roberto Pereira. USDA-ARS, CMAVE
8:48	9:00	<b>Establishment of <i>Pseudacteon curvatus</i> in Mississippi.</b> James T. Vogt and Douglas A. Streett. USDA, ARS BCPRU
9:00	9:12	<b>Involvement of the Extension system in South Carolina Areawide RIFA Suppression Program.</b> Tim Davis. Clemson University Extension; Mac Horton. Clemson University. ( <i>Abstract not submitted.</i> )
9:12	9:24	<b>Mississippi Area-Wide Program: Unique Aspects of Working with Black and Hybrid Imported Fire Ants.</b> James T. Vogt and D.A. Streett. USDA, ARS, BCPRU, Stoneville, MS; Roberto M. Pereira. USDA, ARS, Imported Fire Ant and Household Insect Research Unit, Gainesville, FL; Anne-Marie Callcott. USDA-APHIS-PPQ, Gulfport
9:24	9:36	<b>Communicating the Benefits of the Areawide Suppression Project.</b> Phil Koehler. University of Florida. ( <i>Copy of presentation included on attached CD</i> )

### **Community-Wide Suppression Project**

9:36	9:48	<b>The Lago Santa Fe Fire Ant Project: Community-wide Imported Fire Ant Management in Texas.</b> Paul R. Nester, Corrie P. Bowen, and Bastiaan "Bart" M. Drees. Texas Cooperative Extension
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### **Environmental Concerns**

9:48	10:00	<b>Mitigation of Bifenthrin in Nursery Runoff.</b> John Kabashima. University of California Cooperative Extension
10:00	11:00	<b>REFRESHMENT BREAK</b> - Featuring date shakes



## Posters and Exhibits

11:00	12:00	<b>Business Meeting</b>
12:00	5:00	<b>Poster Session</b>
5:00		<b>Adjourn</b>

## List of Poster Presentations

1. **The Ants Underground: Youth CD-ROM.** D Shanklin, T Riley, C Meux, and J Shivers. University of Arkansas, Cooperative Extension
2. **Cindy Heights Neighborhood Abatement Projects in Jefferson CO Arkansas.** J Ivy, D Shanklin, K Loftin, J Hopkins. University of Arkansas, Cooperative Extension Service
3. **Evaluation of Broadcast Applications of Various Contact Insecticides.** J Gavin, K Loftin, D Shanklin, J Hopkins. University of Arkansas, Cooperative Extension Service
4. **Biological Control Releases in Arkansas 2002; *Psuedacteon tricusps* and *Thelohania solenopsae*.** K Loftin, M McCarter, D Petty, J Gavin, J Clemons, J Hopkins, D Shanklin. University of Arkansas Cooperative Extension Service; J Bard. USDA-PPQ
5. **A List of the Ants of Mississippi with a Key to the Genera.** T Lockley. USDA-APHIS-PPQ-SIPL
6. **Morphological Embryonic Development of RIFA.** Xing Ping Hu, Yongyu Xu & Michael Williams. Auburn University; T Lockley. USDA-APHIS-PPQ-SIPL
7. **Release and Establishment of the Fire Ant Decapitating Fly, *Pseudacteon tricusps*, in the Southeastern United States.** Sanford D. Porter. USDA-ARS; Lawrence E. Gilbert. U of Texas; Seth J. Johnson. Louisiana State U; Lawrence C. Fudd-Graham. Auburn U; Paul M. Horton. Sandhills Research & Education Center; and Tim Lockley. USDA, APHIS
8. **Potential Global Range Expansion of the Invasive Fire Ant, *Solenopsis invicta*.** Lloyd W. Morrison, Sanford D. Porter & Eric Daniels. USDA-ARS, Center for Medical and Veterinary Entomology, Gainesville, FL; and Michael D. Korzukhin. Institute of Global Climate and Ecology, Moscow, Russia
9. **Identification of Polygyne and Monogyne Fire Ant Colonies (*Solenopsis invicta*) by Multiplex PCR of Gp-9 Alleles.** Steven M. Valles and Sanford D. Porter, USDA-ARS, Center for Medical and Veterinary Entomology, Gainesville, FL
10. **State-Wide Distribution of Imported Fire Ant Populations in Mississippi.** D.A. Streett. USDA, ARS, Biological Control of Pests Research Unit, Stoneville, MS; Thomas Barton Freeland, Jr. Mississippi State University, Delta Research and Extension Center, Stoneville, MS; Robert K. Vander Meer. USDA, ARS, Imported Fire Ant and Household Insect Research Unit, Gainesville, FL
11. **Mississippi Area-Wide Management of Black Imported Fire Ants on Pasture.** D.A. Streett and James T. Voat. USDA. ARS. Biological Control of Pests Research



Unit, Stoneville, MS; Roberto M. Pereira. USDA, ARS, Imported Fire Ant and Household Insect Research Unit, Gainesville, FL

12. **Preliminary assessment of *Dorymyrmex* and *Solenopsis* interactions.** Alejandro Calixto, Marvin K. Harris, Allen Knutson, and Charles Barr. Department of Entomology, Texas A&M University
13. **Areawide Suppression of Fire Ants: Texas.** Charles Barr, Alejandro Calixto and Bart Drees. Department of Entomology, Texas A&M University
14. **The Use of Community Participation in Surveillance for RIFA in SouthEast QLD, Australia.** Michelle Milzewski, Jenny Bibb. Australia Fire Ant Control Centre
15. **The Use of Public Relations Activities in Passive Surveillance for RIFA in South East QLD, Australia.** Jenny Bibb, Michelle Milzewski. Australia Fire Ant Control Centre
16. **Diagnostics & Its Role in the Fire Ant Eradication Program in Brisbane. Australia.** Marlene Elson-Harris, Kym Johnson, Shane Moloney, and Lynne Griffin. Australia Fire Ant Control Centre
17. **Increasing the Treatment Options for Fire Ants in Brisbane, Australia.** John R. Hargreaves and John Johnston. Australia Fire Ant Control Centre
18. **Progress of the Fire Ant Eradication Program in Brisbane Australia.** Craig Jennings. Australia Fire Ant Control Centre
19. **Ecological effects of *Solenopsis invicta* in Brisbane Australia.** Tania Fuessel, Adriana Najar, Jo-Anne Holley, and Kris Plowman. Australia Fire Ant Control Centre
20. **Monitoring the Fire Ant Eradication Program in Brisbane Australia.** Evan Harris, Stuart Mutzig, and Paul Garland. Australia Fire Ant Control Centre
21. **Pastoral Peace? A Third Year Report on *Thelohania solenopsae* in a Mississippi Coastal Pasture.** Shannon James. USDA, APHIS, PPQ, CPHST, SIPL
22. **A Simulation Model of Competitive Interactions Among Polygyne Imported Fire colonies for Foraging Space and Food Resources.** R.D. Weeks, L.T. Wilson, S.B. Vinson, and M.J. Yoder. Dept. of Entomology, Texas A&M University
23. **Endocrine and Ovarian Changes in Newly Dealate Queens of *Solenopsis invicta*.** Colin Brent and Ed Vargo. Dept. of Entomology, North Carolina State University
24. **Distribution Patterns of *Thelohania solenopsae* Spores in the RIFA Mounds in Southern Oklahoma.** Vedham Karpakunjaram, Wayne A. Smith and Russell E. Wright. Oklahoma State University
25. **Eradication Strategies in an Orange County, Ca. Neighborhood.** Charlie Cassidy & Shana Low. Orange County Fire Ant Authority
26. **Degradation of Bifenthrin and Deltamethrin on Several Substrates.** Caren L. Carney, Linda M. Hooper-Bui, Jessica Rosson, and Heather Whitney. Department of Entomology, Louisiana State University
27. **Effects of Red Imported Fire Ants *Solenopsis invicta* Buren on Soil Chemistry in Louisiana.** Laura Allen, Linda M. Hooper-Bui, and Benoit Lafleur. Department of Entomology, Louisiana State University

28. **Influence of Phorid Flies and Low Humidity on Foraging Strategies of *Solenopsis* spp.** Ricardo A. Ramierz II, David C. Thompson(\*), and Marta D. Remmenga. New Mexico State University
29. **Red Imported Fire Ant Management in a South Louisiana Citrus Orchard.** Dale Pollet, Patricia Beckley, Bobbie Fletcher, and Boris Castro. Louisiana State University AgCenter Entomology Department
30. **Seasonal Effects of Temperature on Red Imported Fire Ants (Hymenoptera: Formicidae).** Vicky E. Bertagnolli, L. C. 'Fudd' Graham, and Arthur G. Appel. Department of Entomology and Plant Pathology Auburn University Auburn, AL
31. **Evaluation of Mechanical Disturbance of Mounds During Cold Weather on Red Imported Fire Ant Populations.** L. C. 'Fudd' Graham. Department of Entomology and Plant Pathology Auburn University Auburn, AL; Vicky E. Bertagnolli, and Amber T. Kelley. Department of Entomology and Plant Pathology Mississippi State University Starkville, MS
32. **Red Imported Fire Ants reduce Lepidopteran pests in cotton but not in soybean.** John D. Styrsky and Micky D. Eubanks. Auburn University
33. **Non-Avoidance of Sodium Bicarbonate-Treated Surfaces and Food by the Red Imported Fire Ant (Hymenoptera: Formicidae).** Mark A. Brinkman, Wayne A. Gardner and Reid M. Ipser. University of Georgia
34. **Performance of Organic Insecticides as Individual Mound Treatments Against Red Imported Fire Ants.** Marla J. Tanley and Arthur G. Appel. Department of Entomology and Plant pathology, Auburn University
35. **Effects of Temperature and Relative Humidity on Water Regulation of Alate Red Imported Fire Ants.** Arthur G. Appel and Marla J. Tanley. Department of Entomology and Plant Pathology, Auburn University
36. **Apparent Facilitation of Ground-Dwelling Mealybugs by the Red Imported Fire Ant, *Solenopsis invicta*.** Ken R. Helms and S. Bradleigh Vinson. Dept. of Entomology, Texas A&M University



## Keynote Speaker

### ***Does *Solenopsis invicta* really suppress native ants?***

Walter R. Tschinkel

Department of Biological Sciences, Florida State University

For half a century of more, it has been widely believed that the fire ant, *Solenopsis invicta*, suppresses populations of native ants. This belief has become an oft-repeated mantra in the scientific literature, yet a careful re-reading of the evidence does not clearly support this claim (except for the two congeners, *S. geminata* and *S. xyloni*). First, the claim is always based on correlations rather than experimental evidence, and second, there is widespread reliance on relative ant abundance data rather than absolute abundance. Reanalysis of several studies showed that whereas relative abundance data gave the appearance that native ants were being suppressed, absolute counts showed that the number of native ants and the number of their species remained constant or went up in the presence of *S. invicta*. In contrast to a direct effect of *S. invicta* on native ants, many studies suggest that native ants are suppressed by ecological disturbance, and that the same disturbance directly favors fire ants, creating a non-causal correlation between *S. invicta* and native ant abundance. Wojcik's 20-year study near Gainesville, Florida confounded the disturbance of rapid urbanization with the invasion of *S. invicta*. Our study of the ants of the piney flatwoods of the Florida coastal plain showed a decline in species richness as the frequency of disturbance by fire and flooding increased. All but the last portion of this decline occurred in the absence of *S. invicta*, and therefore must be attributed to disturbance, rather than *S. invicta*. Another confusion in this literature is caused by the failure to distinguish between social forms of *S. invicta*. The study most commonly cited to support the suppression of native ants by monogyne *S. invicta* is that of Porter et al. on the invasion of Brackenridge Field Station near Austin, Texas. Such use of this citation is inappropriate because the invader was the polygyne form of *S. invicta*, a form with a very different biology than the monogyne form. In any case, resurvey of the Brackenridge site ten years later showed that all the native ants (except *S. geminata* and *Pogonomyrmex*) had rebounded to their pre-invasion levels.

Altogether, the published data suggest that with respect to the suppression of native ants, ecological disturbance plays a central role, and *S. invicta* has little direct effect. Experimental studies are desperately needed.

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- 44      **Phorid Flies in Alabama: A Tale of Two Species.** L. C. 'Fudd' Graham. Department of Entomology and Plant Pathology Auburn University Auburn, AL; Sanford.D. Porter. USDA-ARS, CMAVE Gainesville, FL; Vicky E. Bertagnolli. Department of Entomology and Plant Pathology Auburn



- 45      **Evaluation of Organic™ Solutions All Crop Multipurpose Commercial and Agricultural Insecticide as a Red Imported Fire Ant Mound Drench Treatment.** Nathan Riggs. Texas Cooperative Extension, San Antonio; Wizzie Brown. Texas Cooperative Extension, Austin; Dr. Bart Drees. Texas Cooperative Extension, College Station, TX
- 50      **Fire Ant Mound and Foraging Suppression by Indoxacarb Bait.** Charles L. Barr. Texas Cooperative Extension, College Station, TX
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**Note: Two speakers (Richard S. Patterson and Phil Koehler) submitted their abstracts as Powerpoint presentations. Their files can be viewed on the attached CD.**

## Temporal foraging activity of selected ant species in northern Mississippi

James T. Vogt, Jack T. Reed<sup>1</sup>, and Richard L. Brown<sup>1</sup>  
USDA, ARS Biological Control of Pests Research Unit, P.O. Box 67, Stoneville, MS

**ABSTRACT** An experiment was conducted in northeastern Mississippi to examine temporal foraging activity of imported fire ants and other common ant species that inhabit pasture and meadow areas. Baited vials were placed horizontally on the ground along straight-line transects ( $N = 21$ ) every 3 h for 24 h periods during June-August. Vials remained on the ground for 30 min, then were quickly plugged with cotton and collected. Principal species captured in baited vials included *Solenopsis richteri* × *invicta* (hybrid imported fire ant) (90.6%), *Solenopsis molesta* (Say) (5.9%), *Monomorium minimum* (Buckley) (2.5%), *Tapinoma sessile* (Say) (0.7%), and *Paratrechina vividula* (Nylander) (0.3%). Imported fire ants foraged during all time periods, as did *S. molesta* and *P. vividula*. *Tapinoma sessile* and *M. minimum* slowed or ceased foraging at night. *Forelius pruinosus* (Roger) was captured on a single date while sampling at 1800 h.

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<sup>1</sup> Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State, Mississippi 39762



## Co-occurrence of ant species with the red imported fire ant in South Carolina

Tim Davis, Paul "Mac" Horton, and Craig Allen  
Clemson University

**Introduction:** The negative impact of the Red Imported Fire Ant *Solenopsis invicta* (RIFA) on native species has long been discussed. At the same time many species do co-occur. The last ant survey conducted in South Carolina was conducted by M.R. Smith and published in 1919. This study predates the occurrence of RIFA. In order to determine the impact of RIFA on native ant fauna it must first be determined which species occur, and in what habitats as well as which species co-occur with RIFA. This report focuses upon the habitats in which RIFA was found to occur in the piedmont and mountains of South Carolina and which species were found to co-occur.

**Materials and Methods:** Ants were sampled by pitfall traps throughout the state of South Carolina, USA. Samples were stratified by the physiographic regions: mountain, piedmont, sandhill, and coastal plain. Sampling efforts were further stratified by land cover types as defined by the South Carolina Gap Analysis Program (SC-GAP).

**Results:** Results are summarized in two tables. In the mountains RIFA were only collected in two of the sampled habitats: dry deciduous forest, and dry mixed forest. 24 and 33 species were found to co-occur with RIFA in these habitats. In the piedmont RIFA were collected in seven of the sampled habitats: Swamps/Bottomland Hardwood forests 31 species, Open Canopy, Recently Cleared Forest 14 species, Needle-leaved Evergreen Forest/Closed Canopy 18 species, Mesic Deciduous Forest 21 species, Dry Mixed Forest 16 species, Mesic Mixed Forest 16 species, and Grasslands/Pastures 20 species.

**Conclusions:** These data clearly represent the species richness of the ant fauna that are capable of co-occurring with RIFA. They do not, however, represent the effect that RIFA may have on the abundance or biomass of co-occurring species.

Future reports will focus on the co-occurrence of RIFA in the sandhill and coastal plain regions of South Carolina as well as the impact of RIFA on the abundance of co-occurring species.

(Tables Attached)

<i>Amblyopone</i> sp	1	0	0	1	1	1	0
<i>Apheanogaster fulva</i>	1	0	0	0	0	0	0
<i>Apheanogaster ashmeadi</i>	0	1	1	1	1	1	1
<i>Apheanogaster mariae</i>	1	0	0	1	0	0	0
<i>Apheanogaster picea</i>	1	0	1	1	0	0	0
<i>Apheanogaster rudis</i> group	1	1	1	1	1	1	1
<i>Camponotus americanus</i>	0	1	0	1	1	0	0
<i>Camponotus pennsylvanicus</i>	1	0	1	1	0	0	0
<i>Crematogaster Cerasi</i>	0	0	0	1	0	0	0
<i>Crematogaster atkinsoni</i>	1	0	0	1	1	1	0
<i>Crematogaster linedolata</i>	1	1	1	1	1	1	1
<i>Crematogaster pilosa</i>	1	1	0	0	0	0	1
<i>Dorymyrmex bureni</i>	1	0	0	0	0	0	1
<i>Formica integra</i>	1	0	0	0	0	0	1
<i>Formica pallidefulva</i>	1	0	0	0	0	0	1
<i>Formica schaufussi</i>	1	1	1	0	0	1	1
<i>Formica subsericea</i>	1	0	1	1	0	0	0
<i>Hypoponera</i> sp	1	0	1	1	0	1	1
<i>Leptothorax</i> spp	1	0	1	1	1	0	1
<i>Monomorium minimum</i>	1	1	0	0	0	0	1
<i>Myrmecina americana</i>	1	1	1	1	1	1	0
<i>Myrmica</i> sp	0	0	0	1	0	0	0
<i>Neivamyrmex opacithorax</i>	1	0	1	1	0	0	0
<i>Neivamyrmex texanus</i>	1	0	0	0	1	1	1
<i>Pachycondyla chinensis</i>	1	0	0	0	1	0	0
<i>Paratrechina arenivaga</i>	1	0	0	0	0	0	1
<i>Paratrechina faisonensis</i>	1	1	1	1	1	1	1
<i>Paratrechina parvula</i>	1	0	0	0	0	0	1
<i>Phidole</i> spp	1	1	1	1	1	1	1
<i>Prenolepis imparis</i>	1	1	1	1	1	1	1
<i>Solenopsis carolinensis</i>	1	1	1	1	1	1	1
<i>Solenopsis Invicta</i>	1	1	1	1	1	1	1
<i>Strumigenys gundlachi</i>	1	0	1	0	1	1	0
<i>Tapinoma sessile</i>	1	1	1	0	0	1	1
<i>Tracymyrmex spetentrionalis</i>	1	0	0	0	0	0	0
<b>number of species</b>	<b>31</b>	<b>14</b>	<b>18</b>	<b>21</b>	<b>16</b>	<b>16</b>	<b>20</b>

Table 1: Presence or absence of species collected and co-occurring in seven land cover types in the South Carolina Piedmont.



<i>Ambylopone</i> sp	1	1
<i>Apheanogaster fulva</i>	1	1
<i>Apheanogaster picea</i>	1	1
<i>Apheanogaster rudis</i> group	1	1
<i>Camponotus americanus</i>	1	1
<i>Camponotus chromoides</i>	1	1
<i>Camponotus pennsylvanicus</i>	1	1
<i>Crematogaster ahmeadi</i>	1	0
<i>Crematogaster cerasi</i>	1	0
<i>Crematogaster atkinsoni</i>	0	1
<i>Crematogaster lineolata</i>	1	1
<i>Crematogaster pilosa</i>	0	1
<i>Formica argenticola</i>	0	1
<i>Formica integra</i>	1	1
<i>Formica schaufussi</i>	1	1
<i>Formica subsericea</i>	1	1
<i>Hypoponera</i> sp	1	1
<i>Leptothorax</i> spp.	1	1
<i>Monomorium minimum</i>	0	1
<i>Myrmecina americana</i>	1	1
<i>Myrmica americana</i>	1	0
<i>Myrmica</i> spp.	1	1
<i>Neivamyrmex opacithorax</i>	0	1
<i>Neivamyrmex texanus</i>	0	1
<i>Paratrechina faisonensis</i>	1	1
<i>Ponera</i> sp	0	1
<i>Prenolepis imparis</i>	1	1
<i>Solenopsis carolinensis</i>	1	1
<i>Solenopsis invicta</i>	1	1
<i>Stenamma</i> spp.	1	1
<i>Stenamma schmittii</i>	0	1
<i>Strumigenys gundlachi</i>	0	1
<i>Tapinoma sessile</i>	1	1
<i>Tracymyrmex spetentrionalis</i>	0	1

24 species    33 species

Table 2: Presence or absence of species collected and co-occurring in two land cover types in the South Carolina Mountains.

## Potential ecological impacts of Red Imported Fire Ants in eastern Australia

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### Abstract

Red Imported Fire Ants are a relatively new arrival to Australia. Currently a concerted eradication program is in place which aims to eradicate them by 2006. Should Red Imported Fire Ants not be eradicated they are likely to spread to many parts of the Australian continent. There can be no doubt that *Solenopsis invicta* will pose a substantial risk to Australia's fauna if it spreads beyond its current Australian range and is not eradicated. If the worst case scenario occurs and their range increases to cover most of the continent as predicted, wide-ranging species declines in a variety habitats are to be expected. While endangered species are of particular concern, many common Australian animal species have experienced range declines, and the additional pressure caused by *S. invicta* may be sufficient to result in a new wave of species losses. It is crucial that we determine which groups have already been negatively affected by fire ants in Australia, and that we establish which fauna is most at risk to ensure any future research and conservation funding is applied appropriately.

(Full article submitted to special issue of Journal of Agricultural and Urban Entomology)



# **Landscape Maintenance Practices Influence Fire Ant Establishment**

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## **Introduction**

The red imported fire ant (RIFA), *Solenopsis invicta* Buren, was introduced into United States near Mobil, AL in the 1920s from Brazil, South America, and now infests ca. 109 million ha (270 million acres) across the Southern United States from Florida/North Carolina to California, Puerto Rico and it is established in several countries of South America, Australia and New Zealand (Drees et al. 1998). The costs for controlling this pest in five large metropolitan areas in Texas in 1998 were estimated at US\$581 million. Additionally, it was estimated that RIFA cost each homeowner US\$151 per year and that expenditures in the five major metropolitan areas in the state exceeded US\$526 million for households, US\$112 million for cities, US\$29 million for golf courses and US\$25 million for schools (Lard et al. 2000).

Although it is a major agricultural pest, it is also considered one of our most important economic pests in the urban landscape. RIFA causes damage by its extensive mounding and tunneling and disrupts recreational activities with its aggressive stinging. RIFA stinging can cause considerable medical problems, with costs estimated to exceed US\$47 million in the 5 larger cities in Texas (Lard et al. 2000). The ants sting repeatedly and attack anything or anyone near the colony when it is disturbed. Additionally, RIFA are attracted to current and electrical equipment (Slowik et al. 1996). Estimated damage to this equipment in the Texas economic study was placed at US\$111 million (Lard et al. 2000). For these reasons, control measures are often necessary in urban landscapes around residential and commercial buildings, in parks and on and around other recreational and sports turf facilities.

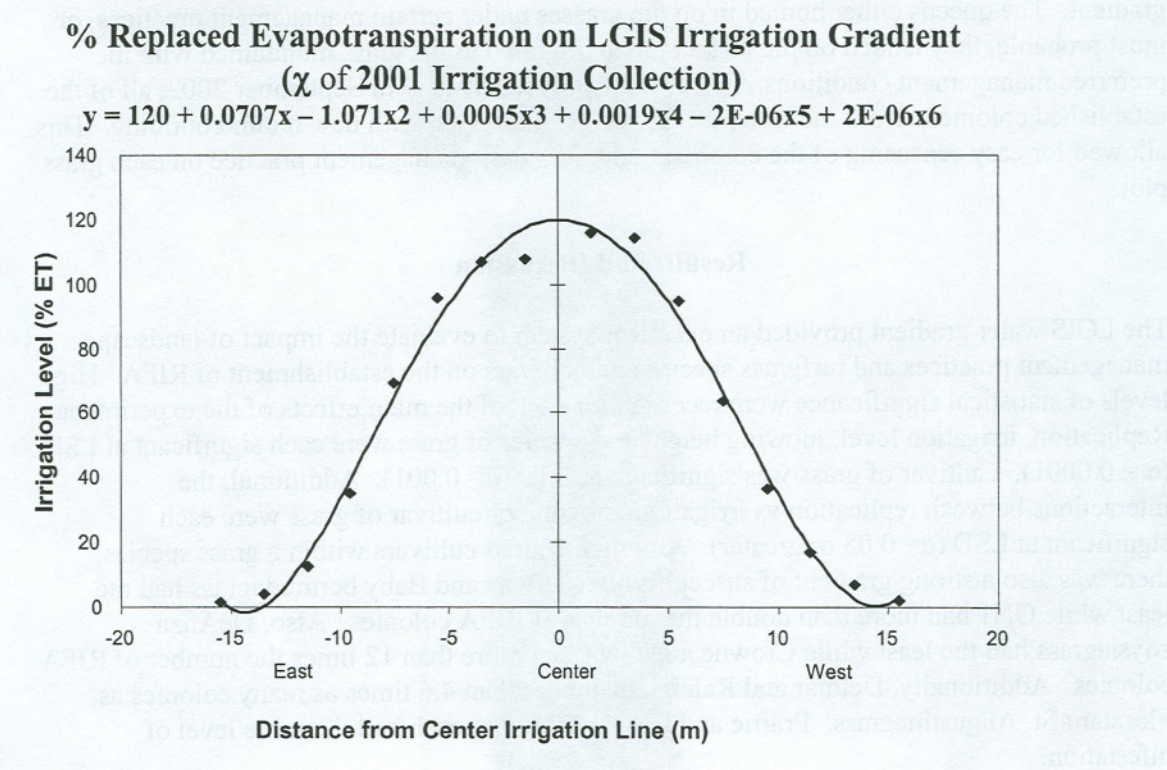
Effective, safe and environmentally sound management strategies must be developed for this serious pest in all aspects of the urban landscape. This study was initiated to determine if RIFA establishment and its densities could be manipulated by changing management practices in the turf/landscape environs.

## **Materials and Methods**

For this study turfgrass plots planted across a Linear Gradient Irrigation System (LGIS) were used to assay the influence of several landscape management practices on RIFA establishment and habitat preference. Management practices evaluated included: a) irrigation across the gradient (high = > 95% replaced evapotranspiration, medium = 30-80% replacement and < 20% replacement), b) mowing height [3 cutting heights = 1.25cm (1/2 inch), 2.5cm (1 inch) and 5cm (2 inch)], c) four grass species (Table 1) and d) 20 turf



**Figure 1.** Distribution of irrigation across turf plots planted on the Linear Gradient Irrigation System. Note: Supplemental water ranged from ca. 120% at center trench to 0% at ca 16 m away.



**Table 1.** Grass species and cultivars evaluated across the Linear Gradient Irrigation System (4 replicates).

Bermudagrass	Zoysiagrass
Tifway	DeAnza
Baby	Jamur
Tifton 10	Cavalier
Tifsport	Zeon
GN1	Compatible
	Palisades
St. Augustinegrass	El Toro
Floritam	Zenith
Delmar	GNZ
Raleigh	Crowne
Buffalograss	
609	
Prairie	

cultivars (Table 1). Each grass was planted perpendicular to the water source allowing the water gradient to extend across each grass plot in each of the four replicates.



After high rainfall during the spring of 1992, we experienced heavy alate flights of RIFA. Large numbers of the mated queens landed all across the turf plots on the LGIS water gradient. The queens either homed in on the grasses under certain management practices, or most probable, they landed on the turf and then migrated to the grass maintained with the preferred management conditions. Following heavy rains again in September 2002, all of the established colonies across the water gradient were expressed with new mound building. This allowed for easy censusing of the colonization under each management practice on each grass plot.

## Results and Discussion

The LGIS water gradient provided an excellent system to evaluate the impact of landscape management practices and turfgrass species and cultivars on the establishment of RIFA. High levels of statistical significance were recorded for each of the main effects of the experiment. Replication, irrigation level, mowing height and species of grass were each significant at LSD ( $\alpha = 0.0001$ ). Cultivar of grass was significant at LSD ( $\alpha = 0.001$ ). Additionally, the interactions between replication vs irrigation, mowing or cultivar of grass were each significant at LSD ( $\alpha = 0.05$  or greater). Among the grass cultivars within a grass species, there was also a strong gradient of susceptibility. Tifway and Baby bermudagrass had the least while GN1 had more than double the number of RIFA colonies. Also, DeAnza zoysiagrass had the least while Crowne and GNZ had more than 12 times the number of RIFA colonies. Additionally, Delmar and Raleigh had more than 4.6 times as many colonies as Floratam St. Augustinegrass. Prairie and 609 buffalograss each had the same level of infestation.

Overall, RIFA appeared to prefer to establish its colonies in the more open turf with lower soil moisture and maintained at the higher mowing level. Figure 2 shows the overall effects of mowing height and irrigation level (soil moisture). Note the higher colony density (each colony is marked with a black dot) in the section of the plot maintained at 5 cm cutting height and the apparent migration of colonies away from the high soil moisture.

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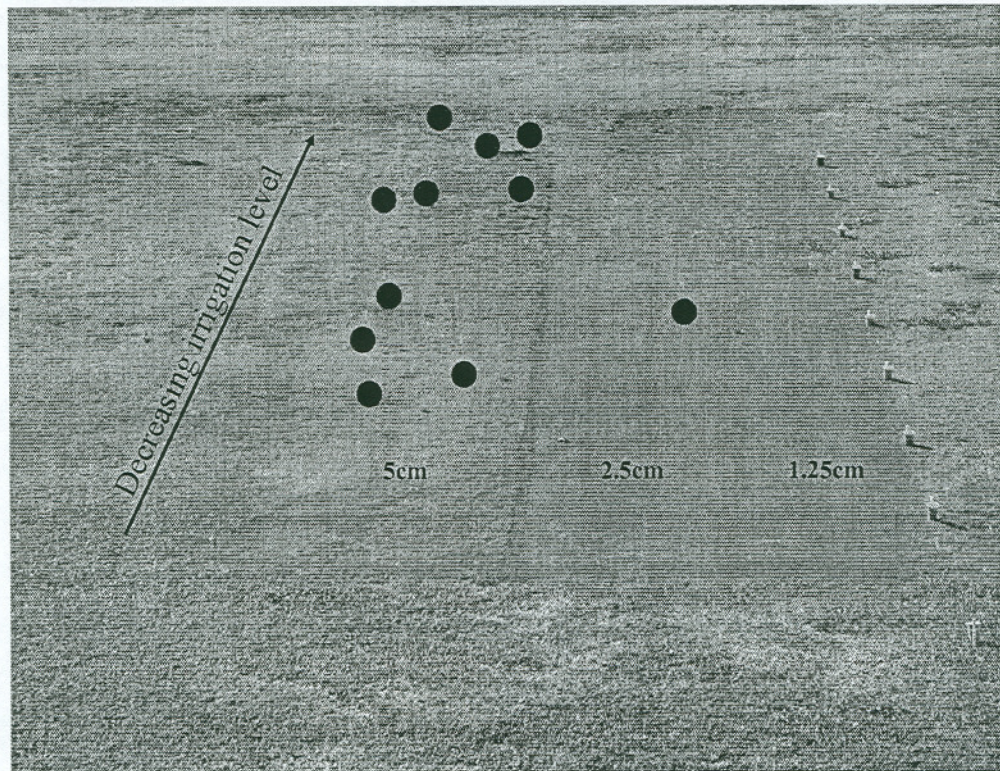
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**Figure 2.** One of the turf plots showing the effect of mowing height and irrigation level (soil moisture). Note the higher number of RIFA colonies in the taller grass and at the lower soil moisture.





# Observations on Fire Ants and *Iridomyrmex* species during an eradication program in Brisbane, Australia

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The red imported fire ant, *Solenopsis invicta* Buren, was found in Brisbane, Australia, in February 2001. An eradication program was quickly implemented during the early stages of the invasion. A pitfall-trapping regime was established to monitor 73 infested sites for the presence of *S. invicta* and the efficacy of the program. Individual ants were identified and the most prolific genus collected was found to be *Iridomyrmex*. It is a common genus found throughout Australia and is considered dominant due to its typically high abundance and aggressive nature. This pilot study was conducted to search for any effects of the *S. invicta* invasion on *Iridomyrmex* populations. These pitfall trap data were analysed by comparing the numbers of *S. invicta* and *Iridomyrmex* individuals collected over 15 months. Ten long-term sites were investigated, five sites that were once heavily infested and five sites with very low numbers of fire ants.

Few *Iridomyrmex* individuals were collected when many *S. invicta* individuals were present at heavily infested sites. There was a statistically significant increase in the numbers of *Iridomyrmex* as the number of *S. invicta* decreased (owing to the treatment). No increase in the numbers of *Iridomyrmex* was observed at sites with very low numbers of *S. invicta*. This suggests that the fire ants may have been suppressing the *Iridomyrmex* population or the ants have established the areas after the *S. invicta* were killed. The data sets revealed a statistically significant effect of the seasons on the number of *Iridomyrmex* individuals collected. No statistically significant effect was observed from the presence of Australian native ants. There were few individuals collected relative to *S. invicta* and *Iridomyrmex*. There were significant differences in the numbers of ants collected at the various sites. This will be investigated further as site composition and landscape scale considerations will form the basis of a further study. The main areas to be investigated are habitat complexity, site history, abiotic conditions and landscape scale ecology.



# Seasonal Effects on Detection of Imported Fire Ant Mounds with Airborne Digital Imagery<sup>2</sup>

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**ABSTRACT** An experiment was conducted to test high-resolution (0.25 to 0.1 m) digital imagery as a detection tool for quantifying hybrid imported fire ant colonies in northern Mississippi pasture. True color and false color infrared images of a 240 ha pasture were obtained with a GeoVantage® Geoscanner imaging system consisting of 4 sensors with 10 nm band-pass filters at 450, 550, 650 and 850 nm. Imagery was obtained in May 2002, Aug 2002, Nov 2002 and Feb 2003. On each sampling date, the entire area was imaged at 0.25 m spatial resolution, and partially imaged at 0.1 m spatial resolution. Plots (N = 20) averaging 0.4 ha each were established within the study area and carefully searched for fire ant mounds by a team of at least 3 workers. Each mound was georeferenced, measured (L, W, H), and assigned a unique number. The percentage of the mound surface obscured by emergent vegetation was visually estimated, and mound activity was assessed by probing the mound and looking for worker ants. A photointerpreter familiar with the general appearance of fire ant mounds examined the images at a scale of about 1: 750 and marked suspected mounds. Actual mound locations were overlaid onto the images to check accuracy of detection. Appearance of mounds was highly variable; generally, mounds in spring, fall, and winter images appeared as light or dark spots of bare soil surrounded by a red (false color infrared composite) or green (true color composite) halo of healthy vegetation. Mounds in the August image generally appeared as dull red (false color infrared composite) or green (true color composite) spots, and were very difficult to see; only  $40 \pm 5\%$  (mean  $\pm$  SE) of active mounds were detected in the false color infrared composite. The greatest percentage of active mounds was detected in May in the false color infrared composite ( $72 \pm 5\%$ ). Increasing spatial resolution of imagery did not result in greater detection in May, but did result in a 38% increase in detection during other sampling periods. False color infrared composites were generally superior to true color composites, and active mounds were detected more frequently than inactive mounds, probably due to the ants' mound building and maintenance activity. Predictive models for likelihood of detecting mounds based on individual mound characteristics have been developed and are being considered for publication elsewhere. Airborne multispectral imagery is a useful tool for detecting imported fire ant mounds in Mississippi pasture; additional work is underway to test and implement this technology in different areas and under different conditions.

I thank B. M. Drees and L. C. Graham for helpful comments on an earlier version of the abstract.

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<sup>2</sup> Mention of trade names or commercial products in this publication does not constitute endorsement by the U. S. Department of Agriculture.



## A Look at the Accomplishments and On-going Research of Select Aspects of the Texas RIFA Program

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The "Texas Imported Fire Ant Research and Management Plan" was initially set up as a 6 year program designed to develop the background for new approaches to IFA management and implement the best available management strategies. Here I am going to focus on some of the research and try to provide an overview of the accomplishments of only two of the various basic research programs ("Reproduction" and "Biological Control") that were supported over the last 6 years. Further, I am going to cover the result of the research and input of a number of people that I will list below. In each program some of the results have been published, some are "in press", "in review", "in preparation", or are being analyzed. I have only included references for the papers published, in press or in review; but refer to those in preparation only in the text.

### 1. Reproduction in the Imported Fire Ant.

Participants include Larry Keeley, Patricia Pietrantonio, Ed Vargo, S. B. Vinson, Fernando Consoli, Debbie Cassill, Indera Kariachen, Danny Lewis, Mie-er Chen, and Y. P. Chen.

Reproduction of fire ants is restricted to the queen, as workers do not develop functioning ovaries. Further, female alates do not lay eggs, although in polygyne colonies virgin female dealates will oviposit haploid eggs that develop into males. We initially examined the hemolymph of reproductives (males and females) to establish a base line for the physical-chemical properties of the hemolymph during their development (Consoli and Vinson, 2002). Among the many changes was a 2.7 fold increase in the free amino acid pool of females after they dealated. There were also changes in the protein profile of the hemolymph during development, particularly in females. Among the changes was the occurrence of vitellogenin (VG) which is a soluble yolk protein precursor synthesized and secreted by the fat body. VG is taken up by the oocyte and is deposited into the egg as vitellin (VN). We wanted to determine when VG was produced and released into the hemolymph. To determine this, hemolymph was collected from alates and queens soon after their mating flight and queens from mature colonies. Dr. Keeley and his group, using non-denaturing and SDS-polyacrylamide gel electrophoresis (PAGE), found that VN was present in eggs as a 350 kDa protein band and was composed of two subunits, a ~181 kDa and an ~171 kDa protein band (Lewis et al., 2001). Although VG was present in alate, virgin females 5 days after their eclosion, oocyte development in these virgin queens was not evident for several weeks. However, VG was evident in newly inseminated, dealate queens and then increased over the next 3 weeks. Oocyte maturation reached a peak from 15-25 days after dealation (Lewis et al., 2002).

Examining vitellogenin gene expression revealed that vitellogenin mRNA synthesis begins before adult eclosion and continues in alates, increasing 30% after mating. However, only the 171 kDa VG (VG-II) was present in pre-eclosion alate females, but disappeared by



day 2 after eclosion. In post eclosion alate females the 181 kDa VG (VG-I) becomes the dominate VG. VG-II reappears at the onset of oocyte formation in mated dealate females (Lewis et al., in press). These results suggest that VG-II may be regulated in a way that regulates egg production and thus plays a key role in oocyte development. But what regulates VG-II is unknown.

Ed Vargo and his groups examined JH titers (a whole body analysis) in dealate queens as they matured and began to produce eggs. They found that egg production and JH content were highly correlated, supporting the view that high JH release by a functional queen was involved in egg production and would inhibit the reproduction of virgin queens by suppressing the activity of their corpora allata and subsequent JH production (Brent and Vargo, in press). However, Keeley and Lewis (unpublished) could not stimulate VG gene expression or enhance VG hemolymph titers through treatment with JH mimics. Thus, the regulation of egg production may be more complex than just JH release and is still unanswered.

Another aspect that is involved in egg production, and possibly its control, is the VG receptor (VGR). VGR in the IFA was found to be a member of the low-density lipid receptor (LDLR) super-family and is most closely related to that of chickens. The VGR gene has been identified and cloned (Chen, M.-E. et al., in review). We have also identified a unique sequence that we are developing into a probe for in situ studies to identify the tissue expression of the gene (Pietrantonio, in prep.). We will also investigate the role of hormones in the regulation of VGR expression and we plan to determine if the VGR is involved in the control of reproduction.

We also initiated studies to define some of the other possible factors that are involved in regulating a queens' reproduction. For example, fire ant queens do not appear to lay eggs continually over the course of several years even when considering the increase in the colonies size over this period, and queens in polygyne colonies each lay fewer eggs, suggesting some possible controls. One issue concerns queens in a polygyne colony. We discovered that there is a dominance hierarchy among the queens (Chen, Y. P. and Vinson, 1999). We have also found that subordinate queens prefer to aggregate around the dominate queen. Through the use of feeding studies using isotopes we found that both the subordinate and dominate queens received more food when aggregated (Kuriachan and Vinson, in prep.). Further, we found that subordinate queens are less likely to survive as single queens than the dominate queen (Cassill et al., in review). We also determined that some subordinate queens can become dominate and replace the dominate queen, if they are separated for a while and the feeding level each receives is reversed (reduced food for the dominate queen and the subordinate queen is well fed). Such results suggest that resources may be a factor that must be considered (Kuriachan and Vinson, in prep.). We also found that as larvae pupate and release their meconium that the workers remove the liquid associated with the meconial pellet. This liquid is then fed to the queen and appears to stimulate her reproduction (Cassill et al., in prep). We are preparing to isolate the responsible factor(s). All of this suggests that resource flow has a major effect on reproduction. To examine this further, we initiated studies to examine food resources. We have first focused on carbohydrates.



We found that a major carbohydrate food source is honeydew from a common group of legless mealybugs (Helms and Vinson, 2002). When locating and recruiting to carbohydrates as a major food source, workers use two different strategies depending on the distance to the carbohydrate source (Martin and Vinson, in review), data that also has implications in regard to the use of baits. These studies have also revealed the maximum distance that worker can forage based on their energy needs (Martin and Vinson, in prep). Resource sharing among polygyne colonies also occurs but appears to be much less than originally thought (Weeks et al., in press).

Over the last six years we have gained a lot of information regarding reproduction in fire ants and factors that sustains their reproductive effort. More importantly we have identified several aspects of the ants' biology that suggest there may be some specific factors that regulate the reproductive effort. We have also developed the tools and identified the genes that may be regulated, setting the stage to identify, isolate and characterize the regulatory factor. Once we know these regulatory factors we will be in a position to manipulate them, and in the process, regulate or "turn off" the reproduction of the IFA.

## 2. Biological Control using native species.

Participants include S.B. Vinson, F. Mitchell, K. Snowden, J. Fuxa, T. Cook, F. Consoli, Y. Sokolva, A. Rao, R. Weeks, T. Wilson, J. Chen, M. Pharr, R. Coulson, M.L. Milke, K. Helms, and P. Mokkarala.

During the past 6 year period we focused on two biological control possibilities. One was to investigate the role that native ants might play in influencing fire ant population levels and to determine if it may be feasible to manipulate native ants in some way to impact the IFA population. The second was to examine the potential of manipulating the disease *Thelohania solenopsae* (Microspora) to manage IFA populations.

a) Native ants. We began by examining some aspects of fire ant and native ant biology. There already was evidence to show that several native ants were effective predators of newly mated queens as they began to search for a good location to nest and begin to develop a nest chamber. One question was to determine what native ants persisted in lightly infested areas and determine where the IFA and native ants might most likely interact. We examined native and fire ant interactions in different ecotones in a post oak savanna (Meegan 2001) and in areas where low levels of fire ants exist (Helms and Vinson, 2001, in review). We found that the greatest ant diversity occurred at the wood- grass ecotone boundary with 17 genera. Other ecotones were much less diverse, from 9 to 5 genera. The IFA was most abundant at the grass- pond and wood- field ecotone (Meegan 2001). We also identified a number of ant genera that had co-existed with the IFA for over at least a 6 year period. We found that there were major differences in the foraging windows of some of these native ants and the IFA (Helms and Vinson, 2001, in review). From these species of native ant we selected four species, along with two common but less objectionable invaders, to determine if they were capable of invading small IFA colonies. We found that 5 of the 6 could invade small worker defended IFA colonies, but as the IFA colonies increased in size they became less vulnerable to invasion. We also found that each genera studied had a different point that, based on the colony size of the IFA, they would not invade (Rao and Vinson, 2002). We (Rao and Vinson, in review) also determined that all 5 genera that invaded were also able to kill the



IFA colony. Although one species, *Forelius* sp., would not invade, they did inhibit small IFA colonies from foraging resulting in their starvation and death (Rao and Vinson, in review). We demonstrated that one species, *S. molesta*, could prevent small IFA colonies from establishing if transported into an area they infested (Vinson and Rao, in review). We also confirmed that some native ants would compete with and invade small IFA colonies in simulated field plots located in a green house (Roa & Vinson, in revision). We also showed that 5 of the 6 native ants we studied as competitors and predators were also susceptible to the baits used to manage the IFA (Rao, in prep.).

The available data suggest that serious efforts should be made to encourage certain native ant species and that we should develop ways to re-introduce the native ants back into areas where they have been lost. We also need to develop more IFA specific management approaches that are not impacting the native ants.

b). Evaluation and development of *Thelohania solenopsae* as a biological control agent of the IFA. *T. solenopsae* was discovered in Florida and Texas populations of the IFA by USDA personnel (Williams et al. 1998). In Texas we initiated a study to determine the extent of the infection, initiated studies to determine how the infection is spread horizontally, and began studies on the biology of *T. solenopsae*. We first developed a way to identify the disease with PCR techniques and developed a way to archive the DNA samples for future studies, if needed (Snowden et al., 2002). We have also compared specific DNA regions of several populations of *Thelohania* across the USA and the results suggest that all the populations are similar (Snowden et al., in review). Using staining techniques and UV-laser microbeam dissection and a laser catapulting system, we have confirmed three spore types (Fuxa and group at LSU). We also compared several of the staining methods of *Thelohania* identification with PCR (Milks et al., in review). Using these techniques we have surveyed the state of Texas and have found a decreasing level of infection of colonies as collections are taken moving from West to East (Mitchell et al., in prep). A similar study in Louisiana is nearing completion (Milks et al., in prep).

We have examined fire ant colonies for possible sources of spores and found the most spores in the meconium and in the midden or dead ant piles outside the nest (Chen J. et al., in review). This suggests that horizontal transmission may be from the meconium to larvae and the queen which concurs with the view of Oi et al. (2001) who demonstrated that both infected larvae and workers are needed to get an infection into another colony. Additional research on transmission is underway. We also examined the hemolymph of non-infected and infected ants and have found a down regulation of several proteins including the vitellogenins in infected queens. Surprisingly no novel proteins were found in the blood (Chen J. et al, in prep.). We have also found that infected alate females tend to fly later in the mating flight and many do not fly resulting in the accumulation of infected females in infected nests (Smith et al., in prep.). Infected alates tend to be smaller and have less oil content (Smith et al., in prep.).

We have also been trying to get *T. solenopsae* into a cell culture. We have tried but have not yet developed an ant cell line. However, using some established insect cell lines we have some promising results and we are proceeding to screen a number of additional cell lines to see if we can get an *in vitro* method to raise large numbers of *T. solenopsae* spores.



Overall we have made good progress in beginning to understand the disease and the data suggests that one of the reasons the disease is not as prevalent as we might like is that the disease is not horizontally transmitted easily. But as in most diseases, as the infective agent increases, the incidence of infection increases. This means that if we could produce and spread infective spores around, we could greatly increase the incidence of the disease and as a result have a major negative impact on the IFA population.

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# Identification of gut bacteria from 4<sup>th</sup> instar Red Imported Fire Ant larvae

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**Abstract** We studied the normal culturable bacteria found in the gut of the 4<sup>th</sup>

instar of the Red Imported Fire Ant, *Solenopsis invicta* Buren. The larvae were dissected under sterile conditions and the guts were removed and homogenized in phosphate buffered saline. After growing gram-positive bacteria from this homogenate, 3 of 6 isolates were identified with high confidence using both lipid and genetic analysis. Two of these bacterial isolates were identified as *Lactococcus garviae* and the third as *Staphylococcus saprophyticus*. The three remaining isolates were identified by lipid analysis as *Enterococcus avium*. However, these 3 isolates had a poor genetic match to that bacterium and may therefore be an unknown species.

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## Claustral Founding by Multiple Newly Mated Queens of Mixed Social Forms

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The polygyne social form (multiple queens) of the red imported fire ant *Solenopsis invicta* is becoming more prevalent within its range in the United States. The increased mound density and worker populations associated with the polygyne form translate into an increased impact on humans. The spread of the polygyne form has been documented to occur through budding from existing polygyne populations, but this does not appear to account for the widespread occurrence of this social form. Monogyne and polygyne social forms of *S. invicta* share the same mating flights, thus there is potential for pleometrotic associations between newly mated queens of the same or different social forms, providing a possible mechanism for the increase of polygyne *S. invicta* colonies. We investigated the ability of newly mated queens of the polygyne and monogyne social forms to claustrally found colonies alone or in groups of five. In addition, we studied whether or not newly mated queens of mixed social forms could successfully found colonies. As expected, monogyne queens did well, with a 55% success rate founding colonies individually and 35% when with 4 other monogyne queens. However, polygyne queens were unable to found colonies individually or in groups with other polygyne queens. In pleometrotic situations with mixed social forms with monogyne queens in the majority, a monogyne queen was always the sole survivor. However, when polygyne newly mated queens were in the majority they became the surviving queens after colony foundation. It appears from these results that pleometrotic colony foundation with mixed social forms can provide another mechanism for the spread of the polygyne form well beyond existing polygyne populations.



# Fire Ant (*Solenopsis invicta*) Acoustic Stridulation in response to Phorid Fly (*Pseudacteon tricuspis*) Attack

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## ABSTRACT

Fire ants possess stridulatory organs and produce acoustic sounds, as do many other ant species, presumably as communication of some sort. Although fire ant stridulations have been recorded, their adaptive significance is poorly understood. There is even disagreement in the scientific literature as to whether ants can detect airborne sounds or only substrate vibrations. Furthermore, reception of these signals probably involves mechanoreceptors (small hairs) on antennae and body surface that transduce the particle velocity component of the airborne sound wave (instead of the pressure fluctuations that are perceived by animals with eardrums).

In order to explore the hypothesis that these ant stridulations might be airborne communications and functional in intra-colony communication, recordings were made in an anechoic chamber of fire ants under attack by a single phorid fly (*Pseudoacteon tricuspis*), an ancestral parasite recently imported to this continent for fire ant bio-control. These recordings were analyzed, depicted as visual sonagrams, and compared with recordings of the same ants before the phorid fly was introduced into their container (control).

Stridulations produced with a phorid fly present were more complex, with more repetitive patterns than the 'control' sounds (sans fly). These results support the hypothesis that fire ant stridulations play a role in intra-colony communication; in this case possibly as a warning call.



# **New targets for disruption of imported fire ant pheromone reception**

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## **Abstract**

Ants use pheromones to identify the colony, signal alarms, mark trails to food, attract workers to brood and to the queen, and bring males and females together for mating. Queen pheromones also may be involved in the maintenance of polygyny and in founding slave-making colonies. In addition, foraging, feeding, and defending the nest depend on detection of general odors and tastes and on detection of kairomones (signals from other species). Clearly, interference with pheromone-based communication in a fire ant colony would be a useful goal for RIFA management.

I am developing a method for discovering red imported fire ant pheromones, using affinity chromatography with the pheromone-binding proteins of the ant. Proteins in the PBP/OBP family, which are abundant in moths and other insect species, are not apparent in fire ant antennae. Two other types of hydrophobic ligand-binding protein---chemosensory protein, and apolipoprotein-III---could be important in fire ant olfaction. These proteins may be useful for identifying pheromones by affinity chromatography.

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## Phorid Fly Releases in Arkansas: County Agents Perspective

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Educating Arkansans about the management of fire ants is an important role of the county agent within the Cooperative Extension Service System. Involvement in the release of biological control organisms is not a typical county agent role.

In 2001, three county agents from selected regions in the state were asked to attend a biological control training session in Gainesville, Florida. The agents attending included Jerry Clemons (Clark Co.), Doug Petty (Miller Co.), and John Gavin (Bradley Co.). At the training we were exposed to the techniques used in site selection, pre-evaluation, release, and post-evaluation of two biological control organisms: the phorid fly *Pseudacteon tricuspis*, and the microsporidia *Thelohania solenopsae*.

By being trained by Sanford Porter at the USDA-ARS lab we hoped to improve the potential for a successful release of *Pseudacteon tricuspis*. The training exposed the difficulties involved in rearing the flies prior to their release in the field and a plethora of other potential problems.

The flies were released in May 2002 in Pike County, Arkansas; a county different from any of the counties agents' involved in the training represented. However, it was because of the training we had received that it determined the county that was initially selected to be a release site for the phorid did not have a sufficient fire ant presence to maximize the potential for success. It was for this reason two other counties Pike and Nevada became involved. Pike was selected because it was a neighboring county of a trained agent, and the untrained agent was interested in learning about phorid releases. Nevada County became involved because the control site is located in the agent's county. . This provided another opportunity to educate additional county agents in techniques used to maximize the potential for a successful release of *P. tricuspis*. .

By having trained individuals in Arkansas we were able to have 2 release sites in 2002. The flies used in the Pike county release were part of an exchange of phorid flies and *Thelohania solenopsae* for a contribution to a USDA-ARS research fund. The Bradley county release was made possible due collaboration with Arkansas' USDA-APHIS personnel who needed trained individuals to assist them in the release of flies supplied them by USDA-APHIS.



The release of the phorid flies has had additional benefits for the Cooperative Extension Service – public relations being the most visible. Arkansas residents receive most of their news from Little Rock-based television stations. During the release process all three of the major networks sent crews to the release site. Local newspapers covered the release also. By having the county agent as the ‘front person’ we continue to promote the high education level of our agents and their expertise in various areas of concern. Youth were also involved in the release and evaluation stages of the project. Overall, the experience of being involved in the release of the phorid fly was extremely positive. The Cooperative Extension Service needs to maintain a high profile, and this program enables us to do that easily.



# Prevalence of the fire ant pathogen *Thelohania solenopsae* in monogyne and polygyne colonies

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## ABSTRACT

*Thelohania solenopsae* is a microsporidian pathogen of imported fire ants that debilitates fire ant queens and can cause reductions in fire ant populations. In field sites in ten southern states that were inoculated with *T. solenopsae*, infections were detected in nine sites. However, sustained infections only occurred in four sites and all appeared to contain polygynous populations. In addition, naturally occurring infections in six states also appeared to be in polygyne colonies. To confirm these observations, we determined the natural prevalence of *T. solenopsae* in three pastures in Florida which had varying ratios of monogyne and polygyne red imported fire ant, *Solenopsis invicta*, colonies. For individual colonies at each study site, social form (i.e. monogyny or polygyny) was determined by examining the genotype of ants at the *Gp-9* locus (Valles and Porter 2003). In the U.S., polygynous *S. invicta* colonies contain individuals with both *Gp-9<sup>B</sup>* and *Gp-9<sup>b</sup>* alleles [genotypes: *Gp-9<sup>BB</sup>*, *Gp-9<sup>Bb</sup>*, *Gp-9<sup>bb</sup>*], while monogyne colonies only have the *Gp-9<sup>B</sup>* allele [genotype: *Gp-9<sup>BB</sup>*] (Krieger and Ross 2002). *T. solenopsae* infection was determined by the PCR method described by Valles et al. (2002), which utilized oligonucleotide primers specific to the *T. solenopsae* 16S rRNA gene.

Study sites located in Alachua, Sumter, and Levy counties, had ratios of monogyne:polygyne colonies of 3:55, 28:22, and 13:43, respectively. *T. solenopsae* infections rates at these respective sites were 78, 42, and 61%. *T. solenopsae* infections were only detected in polygynous colonies (100 of 164 total colonies infected [61%]). Of the polygynous colonies, 83% (100/120) were infected. None of the 44 monogyne colonies for all three sites were infected with *T. solenopsae*. A chi-square test of independence over all study sites indicated that the rate of *T. solenopsae* infection was not independent of *S. invicta* social form ( $\chi^2 = 93.96$ ,  $df=1$ ,  $P < 0.001$ ).

While *T. solenopsae* was only detected in polygynous colonies in the field, *T. solenopsae* infections were found in ants with the monogyne genotype. Four colonies reared from field collected, newly-mated queens that were naturally infected with *T. solenopsae*, were found to exhibit the monogyne social form. Thus, *T. solenopsae* infections can occur in genotypes of both social forms. Possible reasons for the high prevalence of *T. solenopsae* field infections in polygynous *S. invicta*, are that infections can be maintained and spread through the exchange of infected brood and the non-synchronous infection of queens in these multi-queen colonies. In monogyne colonies, which are territorial, there is less opportunity to spread infection via brood exchange, and colonies with a single, infected queen die faster than infected polygynous colonies (Oi and Williams 2002). Thus, there is a lower probability of detecting infected monogyne colonies in the field.



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## Phorid Flies in Alabama: A Tale of Two Species

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### Abstract

Two species of phorid fly have been released at 11 sites in Alabama and have been recovered from nine of these sites. *Pseudacteon tricuspidis* was released in South Alabama in populations of the red imported fire ant, *Solenopsis invicta*, and *Pseudacteon curvatus* was released in North Alabama in hybrid fire ant populations (*Solenopsis invicta* x *Solenopsis richteri*). Number of mounds per acre and mound size was recorded for all release sites and for control sites, if established. Data from the three oldest sites are presented. Mound numbers decreased at two release sites, but increased at the third. Further analysis and data collection are needed to determine the long term effect of phorid flies on Alabama fire ants.

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# Evaluation of Organic™ Solutions All Crop Multipurpose Commercial and Agricultural Insecticide as a Red Imported Fire Ant Mound Drench Treatment

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The red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae), is a major economic pest in Texas. Surveys conducted by the Department of Agricultural Economics and the Texas Agricultural Statistics Service have estimated the total annual fire ant damages and expenditures for Texas to be 1.2 billion dollars.

**Objective:** This trial evaluated five currently available mound treatments to control red imported fire ant colonies in a controlled, replicated field test.

## Materials and Methods

This trial was established on the Ingram Ranch in northern Comal County approximately 45 miles north of San Antonio, Texas on November 1, 2002. The treatment area was located at the eastern end of an abandoned pecan orchard on the ranch. The orchard was fenced, preventing entry by livestock, but still accessible to whitetail deer and wild turkeys native to the ranch.

Five currently available products used to control red imported fire ant colonies evaluated and compared are as follows:

1. Organic™ Solutions All Crop Multipurpose Commercial and Agricultural Insecticide (0.2% pyrethrins, 1.0% piperonyl butoxide or PBO and 82.9% silica dioxide) - 4 Tbsp (scoops)/gal water.
2. Bayer® Advanced Lawn® Fire Ant Killer (1.0% cyfluthrin) - 1 tsp or 2 tsp for mounds exceeding 12 inches diameter (only one in this trial), followed by drenching mound with 1 gal water. Note: 10 oz container treated about 65 mounds.
3. Ortho® Fire Ant Killer Granules II (0.2% bifenthrin) - 1 cup/mound, followed by drenching mound with 1 gal water. Note: 3.5 lb bag treated 9 mounds. “Prevents new mounds from forming for up to 4 months.”
4. Terro® Fire Ant Killer (0.05% deltamethrin) - 1 Tbsp/mound applied as dry dust. Note: 24 oz. container treated 45 mounds.



5. Surrender® Brand Fire Ant Killer (75% acephate) - 2 tsp/mound applied as dry dust.  
Note: 16 oz container treated about 108 mounds.
6. Untreated control (wet) using 1 gal water drench per mound
7. Untreated control (dry) left dry.

In addition to the above treatments, soil samples were collected at pre-determined intervals from the Bayer®, Ortho® and Organic™ Solutions groups to determine pesticide residue levels over the period of the trial (see methods described below). This trial was designed to observe the treated mounds over a period of 30 days.

Twenty-eight plots of similar width, but varying in length, each containing ten imported fire ant mounds, were established on November 1, 2002. Plot numbers were arrayed from longest to shortest and divided into four blocks or replicates of seven plots each, with one replicate containing the longest plots, one containing the shortest plots and the remaining, intermediate blocks. Plot corners were marked with colored flags and mounds in that plot were marked with the corresponding color of flag. This method assured that re-invasion of ant colonies migrating from outside plots would be similar for all treatments. Treatments four and five were applied dry without a water drench and the remaining treatments were applied dry followed by an application of one gallon of clean water poured through a colander to simulate a sprinkling can.

At 2, 7, 14 and 30 days after treatment, ant mounds were disturbed slightly and evaluated for ant activity, if any. If a dozen or more worker ants emerged from the slightly disturbed mounds within 30 seconds, the colony was determined to be active. On days 7 and 30 following treatment, the number of "new" colonies appearing within the treatment plots was assessed to determine whether treated ant colonies had moved to new locations or split into more than one colony.

To collect soil samples for pesticide residue analysis, three plots of 20 mounds each were laid out separately from the treatment area and treated in the same manner with either the Bayer®, Ortho® or Organic™ Solutions product on the same day as the rest of the trial. An untreated plot of 20 mounds was also laid out. Prior to treatment and within 1 hour after treatment, soil samples of approximately 10 oz. (285 g.) were collected from each of the four plots described above, placed into a labeled soil sample bag and then placed into a cooler full of ice. In addition, samples were then collected from different mounds within the same plot in a similar manner on day 7, 14 and 30 after treatment. Soil samples were collected at an average depth of four to six inches (10 to 15 cm) from within the sampled mounds. Soil samples were stored in a darkened freezer at 0°F (-17.8°C) until the end of the trial. At the end of the trial, all soil samples were hand-delivered in an ice chest to Environmental Laboratory Services in Austin, Texas, for analysis. During the trial, the median daily temperature was 55°F (12.8°C), the site received 1.2 inches (3.0 cm.) of rain and the soil type at the site was a heavy clay loam vegetated with native bunch grasses and bermuda grasses.

## Results

All treatments significantly reduced ant activity in treated mounds relative to mounds in the untreated check plots (**Table 1.**). At the 2 day mark after treatment, insecticides applied with a water drench were significantly more effective than those applied dry, but all treatments were equally effective by day 14 after treatment. It should be noted that all treatments applied dry did not result in complete elimination of active mounds at any time



during the trial (maximum of 97% reduction), while all treatments applied with a water drench had 100% reduction in activity of treated mounds at the end of the trial. Water applications to the untreated mounds did not result in statistically significant differences in the number of active mounds compared to untreated fire ant mounds left dry, but the disturbance of the untreated mounds due to adding the water drench did result in slightly lower means than those untreated mounds left dry. On the Day 14 and Day 30 evaluations of “new” mound formation in all plots, new mounds were noted in the vicinity of inactive, untreated mounds, especially those that had been “watered.”

Analysis of soil samples (**Table 2.**) revealed that cyfluthrin levels remained highest at the end of the trial while bifenthrin levels were lowest. Pyrethrin levels on average were lowest throughout the trial.

## Discussion

Organic™ Solutions All Crop Multipurpose Commercial and Agricultural Insecticide (0.2% pyrethrins, 1.0% piperonyl butoxide or PBO and 82.9% silica dioxide (amorphous form harvested from freshwater diatoms)), contains plant-derived or botanical ingredients considered by some to be “organic.” Piperonyl butoxide is extracted from the South American *Ocotea* (sassafras) tree, reacted with butylcarbityl and added as a synergist for pyrethrins. There are mixed opinions on the status of PBO as “organic” because of the chemical reaction involved in the extraction process. Regardless, Organic™ Solutions is formulated with botanical pyrethrins extracted from the powdered, dried flower head of the African-grown chrysanthemum, *Chrysanthemum cinerariaefolium*, in contrast to the synthetic active ingredients contained in other products evaluated in this trial. Bifenthrin, cyfluthrin and deltamethrin are pyrethroid insecticides which differ from pyrethrins because they are synthetically produced molecules that have relatively longer persistence when applied to the soil. Soil persistence documented through soil analysis bears this out except on Day 30, when sampling procedures used may have affected the amount of bifenthrin in the treated soil (**Table 2**). Acephate is an organophosphate insecticide which breaks down relatively quickly when in contact with organic material in soils.

Toxicological properties of treatments. Factors to be considered when selecting a product for treating imported fire ant mounds are discussed in fact sheet FAPFS036 posted on the web site, <http://fireant.tamu.edu>. Soil persistence can be a selection factor and in certain instances when products with less soil persistence are desirable. The mode of activity of natural pyrethrins and synthetic pyrethroids is similar (they destabilize nerve cell membranes and are relatively quick-killing). However, other toxicological properties and cost considerations may also be important.

As active ingredients, pyrethrins are relatively less toxic than pyrethroid insecticides (**Appendix 1**). Conversely, when formulated and directed for use, toxicity of applied material can be dramatically changed. Both the toxicity of applied material and the amount of material used in the treatment are additional variables affecting insecticide “load” that results in the environment. Because of these factors, making direct comparison between products when selecting the “least toxic” materials can be difficult.



## Acknowledgements

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**Table 1.** Number of active red imported fire ant mounds/10 mounds treated or per plot following treatment with selected ant mound insecticide product treatment, Nov. 2002, Comal Co., TX.

<u>Treatment</u>	<u>Active ant mounds/10 mounds/plot (4 replications) or total/plot*</u>					
	<u>Day 2</u>	<u>Day 7</u>	<u>Total</u>	<u>Day 14</u>	<u>Day 30</u>	<u>Total</u>
Untreated control (dry)	9.75a	9.75a	10.50a	9.75a	9.75a	12.50a
Untreated control (wet)	8.25a	9.25a	9.25a	9.75a	8.50a	9.75a
Organic™ Solutions (pyrethrins, PBO + DE) – solution (97.4%)	0.25c	0.00c (100%)	0.75cd	0.00b (100%)	0.00b (100%)	2.00c
Bayer® Advanced Lawn® (cyfluthrin) - watered in	0.00c (100%)	0.00c (100%)	0.75cd	0.00b (100%)	0.00b (100%)	2.00c
Ortho® Granules 2 (bifenthrin) - watered in	0.00c (100%)	0.00c (100%)	0.25d	0.00b (100%)	0.0b (100%)	3.00bc
Terro® Fire Ant Killer (deltamethrin) – dry	3.00b (69.2%)	1.75b (82.1%)	2.50b	0.50b (94.9%)	0.25b (97.4%)	2.75bc
Surrender® Brand (acephate) – dry	2.50b (74.4%)	0.25c (97.4%)	2.25bc	0.25b (97.4%)	0.25b (97.4%)	4.25b

\*Means (average values) in columns followed by the same letter are not significantly different using analysis of variance (ANOVA) using Duncan's Multiple Range test at the 0.5 level of probability. Percent controls are in parenthesis under the mean values.



**Table 2.** Soil analysis conducted by Environmental Laboratory Services, Austin, TX, on soil samples from the Ingram Ranch, Nov. 2002, Comal Co., TX.

Insecticide	Day 0 Pre-Trt	Day 0 Post-Trt*	Day 2 Post-Trt	Day 7 Post-Trt	Day 30 Post Trt
pyrethrins <sup>o</sup>	ND <sup>a</sup>	170	140	140	100
bifenthrin	ND <sup>a</sup>	4000	1200	480	45
cyfluthrin	ND <sup>a</sup>	5800	2300	1700	1200

<sup>a</sup> ND - Not Detected at the Reporting Limit

\*Values listed in ug/kg

<sup>o</sup> Organic™ Solutions product contained 0.2% pyrethrins formulated with piperonyl butoxide and silica dioxide or diatomaceous earth, Ortho® product contained 0.2% bifenthrin, Bayer® product contained 1.0% cyfluthrin.

**Appendix 1.** Toxicological properties of Imported Fire Ant Insecticide active ingredients and formulated products (from Agricultural Chemicals, Book I, Insecticides. W.T. Thompson 2001, Product MSDS's).

#### Botanicals

pyrethrins - CAUTION; (rat) Oral LD<sub>50</sub> : **1,500 mg/kg**

Organic™ Solutions Fire Ant Killer (rat) Oral LD<sub>50</sub> : **>5,000 mg/kg**

#### Pyrethroids

bifenthrin - WARNING; Class II; (rat) Oral LD<sub>50</sub> : **54.5 mg/kg**

Ortho® Fire Ant Killer Granules II (rat) Oral LD<sub>50</sub> : **>5,000 mg/kg**

cyfluthrin - CAUTION; (rat) Oral LD<sub>50</sub> : **500 mg/kg**

Bayer® Advanced Lawn® Fire Ant Killer (rat) Oral LD<sub>50</sub> : **3084 mg/kg**

deltamethrin - WARNING; (rat) Oral LD<sub>50</sub> : **128 mg/kg**

Terro® Fire Ant Killer Dust Oral LD<sub>50</sub> : **>5,000 mg/kg**

#### Organophosphates

acephate - CAUTION; Class II; Tech (rat) Oral LD<sub>50</sub> : **947 mg/kg**

Surrender® Brand Fire Ant Killer (rat) Oral LD<sub>50</sub> : **1030 mg/kg**



# Fire Ant Mound and Foraging Suppression by Indoxacarb Bait

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## Abstract

A new bait product containing the active ingredient indoxacarb (DuPont) was used in two field trials to test its effectiveness against red imported fire ants (*Solenopsis invicta* Buren). When broadcast at a rate of 1.1 kg/ha, the material was found to eliminate ant activity in over 90% of mounds within three days during the summer test. Control of mounds during the fall test was much slower, but reached a similar level. However, ant foraging was suppressed to less than 10% of the untreated plots within two days. Such rapid foraging suppression suggests that this bait may provide an alternative to traditional broadcast sprays of contact insecticides, as well as colony elimination that is much faster than any broadcast bait currently available.

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# Repellency and Toxicity of Mint Oil Granules to Red Imported Fire Ants

(Hymenoptera: Formicidae)

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**ABSTRACT** Repellency and toxicity of 2% mint oil granules were evaluated against worker red imported fire ants, *Solenopsis invicta* Buren, in a series of laboratory. In continuous exposure experiments, LT50 values ranged from 1.2 h with 164.8 mg/cm<sup>2</sup> of mint oil granules to 15.3 h with 1.65 mg/cm<sup>2</sup> of granules. LT50 values declined exponentially with increasing density of mint oil granules. Limited exposure to 164.8 mg/cm<sup>2</sup> mint oil granules resulted in >50% KD after 30 min, however there was no KD at 15 min. Twenty-four h mortality increased linearly with increasing exposure time. Mean repellency of worker red imported fire ants ranged from  $49.2 \pm 5.4\%$  for 0 mg/cm<sup>2</sup> (untreated control) of mint oil granules at 30 min to 100% for 147.8 mg/cm<sup>2</sup> of mint oil granules at 3 h. Repellency increased with increasing density of mint oil granules and exposure time.

Mint oil granules could provide another IPM tool for red imported fire ant management, particularly in situations that conventional insecticides would be inappropriate.



# **An update of the Red Imported Fire Ant Program for the California Department of Food and Agriculture**

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## **Abstract**

Initial infestations of Red Imported Fire Ant (RIFA) [*Solenopsis invicta* (Buren)] in the Central Valley were detected in 1997 and were traced back to beehives brought from infested southern states for almond orchard pollination. California Department of Food and Agriculture (CDFA) maintains a dual approach of preventing the entry and spread of RIFA in California through border station inspections, pest interceptions, and through early detection within the state. Following the detection of RIFA in nurseries, landscaped areas, parks, etc in Orange, Riverside, and Los Angeles counties in 1998, CDFA established quarantines in the entire county of Orange and parts of Riverside and Los Angeles Counties. The quarantine was designed to contain RIFA spread by requiring inspection and treatment of articles thought to spread RIFA such as nursery stocks, soil, sod/turf grass, hay, landscaping, and bee colonies.

The Secretary of Agriculture based on recommendations of Science Advisory Panel proposed RIFA Action Plan in March 1999. RIFA Control/Eradication Program was set up by CDFA to control the spread and eradicate known infested sites. The Program, in collaboration with California County Agricultural Commissioners and other agencies cooperatively, survey; treat; and regulate the movement of regulated articles both within and out of the quarantined areas, through inspections, compliance agreements, and certifications. There had been no major RIFA infestations in the state since the quarantine was established. Local Agencies are playing a major role in detection, eradication and public outreach in urban areas. Department of Pesticide Regulation and University of California researchers along with other agencies do environmental monitoring of pesticide use and research.



## **Eradication Strategies in Orange County, CA – A Neighborhood Plan**

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### **Abstract**

In October of 1998, nursery stock originating from Trabuco Canyon, CA was discovered infested with red imported fire ants (RIFA) [*Solenopsis invicta* (Buren)] when delivered to a Las Vegas, Nevada nursery. The California Department of Food and Agriculture (CDFA) quickly responded by surveying the nursery and surrounding neighborhood. They discovered the nursery, an adjoining nursery as well as the surrounding neighborhood was heavily infested with RIFA. With further extensive survey CDFA discovered two major infestations in the north and south parts of Orange County. Quarantines were established in the entire county of Orange along with parts of Los Angeles and Riverside Counties to limit the spread of RIFA in nursery stock, soil, landscaping, hay and bee colonies.

In March of 1999 a RIFA Action Plan was proposed by the Secretary of Agriculture to control/eradicate infested sites and control spread of RIFA. CDFA contracted with California County Agricultural Commissioners and local agencies to cooperatively survey, treat and control the movement of RIFA on the local level. In January 2000 CDFA contracted with the Orange County Agricultural Commissioner's office, which in turn subcontracted with the Orange County Vector Control District. (OCFAA) The Orange County Fire Ant Authority (OCFAA) was established by the vector control district to eradicate RIFA from Orange County.

### **Methods and Materials**

**Outreach-**Through the use of extensive outreach to the infested communities and the county at large the citizens of Orange County were made aware of the threat from red imported fire ants. Partnering with the media allowed OCFAA to reach hundreds of thousands of the people at minimal cost to the program. Newspaper articles, television news spots and local cable television interviews helped to spread the word about RIFA. Citizens were trained to recognize the ants and encouraged to report them to the state's Exotic Pest Hotline number. The number was changed to 1-888-4FIREANT to make it more memorable.

Outreach continued by attending community events and local fairs where people were given information and instructed on how to sample for ants on their own property with the "Pest Test". Public works, utilities and landscaping companies were educated on the dangers of RIFA and given easy reporting methods when they were found. Self addressed mailing logs were given to these groups since they would be on the front lines in the field where they would see the ants first. Flyer racks with information have been distributed to city halls, libraries and other public locations to get the RIFA message to the public.

Finally, staff involvement within each infested neighborhood has been one of our most effective means to educate the people of Orange County about RIFA. Treatment and survey



notices go on every door of every property we treat and survey. Citizens in these neighborhoods have come to know and trust OCFAA staff from the information they distribute and their courteous professional manner.

**Survey-**Original visual and bait surveys of the RIFA infestation by CDFA staff showed two main geographic areas of concern. The northwestern cities of the county contained about 25% of the countywide infestation. The majority of the infestation was in the southeastern communities from Trabuco Canyon south.

OCFAA staff began by using visual survey of large areas to further delimit the spread of RIFA. Staff soon discovered that this method was not sensitive enough to discover small colonies that had not become visible. Visual survey would still be important in neighborhoods where infestations had been found and would be accomplished while treatment and survey notices were distributed to property owners.

Bait surveys using processed lunchmeats and other materials were initiated on sensitive areas of concern such as schools and parks. Since the use of lunchmeats was extremely manpower intensive other options were explored. Various oils including olive, peanut, corn, and canola were tested for RIFA acceptance. All were taken by RIFA at acceptable levels but distribution was still a problem. Common potato chips in sample baskets were used the first year of the program as a sample media but broken chips would fall through the baskets at too large a rate. On staff recommendation corn chips were tried and proved to be the answer to our problem. Presently staff produces over 5000 corn chip baited baskets each week. Delimitation surveys place flags at 50-foot intervals on all areas of potential RIFA colonization. (Approximately 25 baits per acre) Post treatment surveys are done at a higher resolution of 25-foot intervals. (50 baits per acre.

High-risk properties like schools, parks, residences, golf courses and irrigated public medians are baited within a 1-mile grid pattern (state grid plane) around existing positive sites. Post treatment monitoring starts after sites have completed the fourth treatment cycle for an area. Sites that are still positive return to the treatment cycle.

**Treatment-**Treatment of red imported fire ants in Orange County is accomplished with a modified two-step process. Amdro®Pro or Seige®Pro Fire Ant Bait (0.073% hydramethylnon) are used for mound treatment at a mid-label rate of 2 Tbs. (1/2 ounce) per mound. Bait granules are distributed with a shaker vial from the outside perimeter of the mound extending to a 4-foot radius from the center while not disturbing the mound. The remaining area on the site is treated with Distance® Fire Ant Bait (0.5% pyriproxyfen) broadcast outside the 4 foot radius of the mounds at a rate of 1.5 pounds per acre.

Small sites of 1 acre or less are treated using hand held garden spreaders. Large acreage is treated with a Maruyama® MD155 backpack blower adapted for granule dispersal. The blower can be mounted on a golf cart for a faster treatment of parks, schools and slopes in neighborhoods. A Honda® Rancher ATV with a Herd® Spreader is used to treat large parcels of 20 acres or large. These mechanized treatment methods allow staff to treat many acres and hundreds of homes in a single day.



Treatment protocol has also been expanded to reach our goal of countywide eradication. Our primary goal is to keep infested nurseries that are treating for RIFA from being re-infested from the surrounding area. Every property able to support RIFA colonization within ¼ mile of the nursery is placed under a neighborhood treatment protocol. In addition, if a neighborhood has 6 positive sites for RIFA within a ¼ mile diameter, the entire area is treated. Although this means OCFAA treats hundreds of properties that have not shown to be positive for RIFA, it keeps pressure on the overall population and increases the likelihood of eradication.

Treatment cycles are maximized to achieve the eradication goal. Each positive site, neighboring sites within 200 feet of positive finds and neighborhood treatment site all receive 4 treatments at intervals of 90 days or more. This allows OCFAA to stay within label guidelines while putting maximum pressure on the RIFA populations.

**Data Collection**-Original data collection used hand drawn maps and paper records of each RIFA site. It soon became apparent that electronic tracking and data collection would be essential. OCFAA began using the hand held Trimble® Geo Explorer GPS units in January 2001. These units allow staff to enter data in the field quickly and efficiently. Data is automatically downloaded each day to computers where it is exported into the database and a Geographic information System. (GIS) The GPS feature tracks location and associates the information collected with each GPS point on a map. GIS lets OCFAA map and track progress, predict dispersal and use staff most efficiently. With over 130,000 records to date this makes a huge amount of data manageable.

**Progress**- Currently, post treatment monitoring has shown that after the completion of four consecutive treatments the remaining positive sites total less than 5%. In certain target areas such as the original infested nursery and surrounding neighborhood OCFAA has shown over 97% control. As the program continues and further pressure can be kept up on existing RIFA populations more extensive countywide survey will need to be done. This continued pressure along with the natural dryness of the region can lead to the eradication of this pest from Orange County and California.



# An Integrated Pest Management Approach for the Imported Fire Ant

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## ABSTRACT

In June, 2000, the USDA/ARS/Center for Medical, Agricultural, and Veterinary Entomology; EPA's Pesticide Environmental Stewardship Program; Department of Defense, U.S. Army Environmental Center, and Center for Health Promotion and Preventive Medicine; South Carolina Army and Air National Guard; Clemson University; the Council of State Governments, Southern Legislative Conference; and Aventis Corporation initiated a cooperative study using integrated management for fire ants in South Carolina. The objective was to determine if an integrated pest management (IPM) strategy using chemicals plus the release of biological control agents would give longer control of red imported fire ant (RIFA) *Solenopsis invicta* populations thus reducing the number of chemical treatments needed to maintain control. The integrated strategy combined the use of the phorid fly, *Pseudacteon tricuspidis*, the pathogen, *Thelohania solenopsae*, and the chemical fipronil. Three 10-acre (4 ha) test sites each with a 4.5-acre (1.8 ha) treatment plot located in the center were established on Fort Jackson U.S. Army base and McEntire, South Carolina Air National Guard base. Site 1 was treated with the chemical only, 0.1% Fipronil granular at 12.5 lbs per acre (0.0125 lbs of active ingredient per acre). Site 2 was the integrated site in which the chemical fipronil was applied at the same rate as at Site 1 and two biological control agents were released in the area surrounding the treatment. Site 3 was used as an untreated control. Evaluations were conducted twice per year (spring and Fall) on 1/8-acre circular subplots inside and outside of each 4.5-acre treatment plots. Active mounds within each sampling area were assessed using the USDA population index rating system. All active fire ant mounds were mapped to document fire ant populations, reinfestation rates and spread of the biological control agents. Eight pitfall traps were set per 1/8-acre circle to assess the diversity of ant populations. For the first year, average reductions in fire ant populations in the integrated site was 97% and 91% in the chemical only site. However, after 2 years and 2 months where the chemical plus the biological control agents were applied, the RIFA populations in the integrated site were reduced by 100% while in the site with the chemical treatment only, there was only 59% reduction. In the untreated control, there was an increase of 51% in RIFA populations. In addition, *Thelohania solenopsae* was found in 79% of the colonies inspected at the integrated site and phorid flies were discovered attacking fire ant workers 3 miles from the initial release site. Finally, native ant populations increased as the RIFA populations decreased in the integrated site.



# Areawide Suppression of Fire Ant Populations in Pastures: Project Update

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## ABSTRACT

The red imported fire ant, *Solenopsis invicta*, is an invasive species that creates serious medical and agricultural problems, damaging many crops and cattle production. Its annual economic impact to the US economy is approximately \$6 billion dollars. The integration of chemical bait pesticides and biological control agents is utilized in an areawide management program for fire ants coordinated by USDA-ARS scientists at the Center for Medical, Agricultural and Veterinary Entomology in Gainesville, FL. Control sites, where no biological control agents were released, and biocontrol treatment sites, where both decapitating flies and the microsporidium *T. solenopsae* were released, have been established in five states (Florida, Texas, Mississippi, Oklahoma, and South Carolina). All sites received chemical bait applications of a 1:1 mixture of hydramethylnon and methoprene baits applied at 1.5 lb per acre.

To evaluate the effect of the treatments on the arthropod biodiversity, pitfall traps were used twice a year. Decreases in fire ant populations have been observed at the different demonstration sites. The decapitating fly (*P. tricuspsis*) has been established in demonstration sites in 3 states (Florida, Texas and South Carolina); *P. curvatus* was established in Mississippi, Florida and South Carolina. The disease *T. solenopsae* has been established in 4 states (FL, SC, TX, and OK), and continues to spread.

The project has a website (<http://www.ars.usda.gov/fireant/>). This new approach to fire ant control may have significant impact on the management of fire ant populations in the future.

(Full article submitted to special issue of Journal of Agricultural and Urban Entomology)



## Establishment of *Pseudacteon curvatus* Borgmeier in Mississippi

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*Pseudacteon curvatus* Borgmeier, a dipteran parasitoid of imported fire ants, was released at two sites in northeastern Mississippi (Clay County; Knox site and Prima site) as part of the Areawide Suppression of Fire Ants Program. For more information on the Areawide program, see <http://www.ars.usda.gov/fireant/> (last accessed June 30, 2003). Briefly, field-collected ants were exposed to phorid fly attack in the laboratory for 1 wk (50 adult flies / colony / day) then returned to their original mounds in the field. Colonies (N = 24 for Knox, N = 20 for Prima) were collected, exposed to ovipositing flies, and released in May-Jun at Knox and Aug-Sep at Prima. To monitor for flies in the field, trays [27.5 cm wide (23.5 cm inner) X 41.75 cm long (38.25 cm inner) X 12 cm deep] (N = 10) were placed on the ground with about 1 g of worker ants in each, and observed periodically for ovipositing flies. In summer and fall of 2002 field-reared flies were recovered at Knox up to 600 m from the release area. Establishment and spread of *P. curvatus* in Mississippi is detailed in Vogt and Streett (2003).

Field-reared flies were recovered at Knox in spring 2003 as far as 2 km from the release area, confirming that the population had overwintered and continued to spread. In extensive sampling at Prima in 2003, no ovipositing flies were recovered and a second release has since been conducted (May 2003). Early season releases may be more likely to succeed than late season releases, as the flies presumably can build up larger population density prior to the onset of winter. Distance between the Knox and Prima sites is about 9.6 km; another phorid species, *P. tricusps*, has been shown to spread at a rate of about 16 to 32 km / yr in Florida (S. D. Porter, personal communication), and *P. curvatus* flies have been collected over 19 km from their point of introduction in central Alabama 2 years after establishment (Graham et al. 2003). Sampling will continue in Mississippi to determine rate of spread and success of introductions.

### Acknowledgments

We thank S. D. Porter (USDA, ARS CMAVE) for providing the source colony of *P. curvatus* for mass rearing in Mississippi. John R. Davis, Larry Thead, Alfred Martin and Evita Gourley provided expert assistance in the laboratory and the field. We thank B. M. Drees and L. C. Graham for helpful comments on an earlier version of the manuscript.

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Mississippi Areawide Program: Unique Aspects of Working with Black and Hybrid  
Imported Fire Ants

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**ABSTRACT** Chemical and biological controls for imported fire ants (*Solenopsis invicta* Buren, the red imported fire ant, *S. richteri* Forel, the black imported fire ant, and their hybrid) are being investigated and demonstrated in a multi-state, multi-agency Areawide Management Program. The Mississippi component of the program offers the unique opportunity to implement the latest control technologies against monogyne black and hybrid imported fire ants, since sites in the other participating states (Florida, South Carolina, Texas, and Oklahoma) are dominated by polygyne red imported fire ants. Several observed and potential differences were examined between populations of black / hybrid fire ants and populations of red imported fire ants. Lower nest density of monogyne black and hybrid fire ant populations (range = 19 to 52% of mean nest density at other sites) required modification of sampling protocols. *Pseudacteon curvatus* Borgmeier, a phorid fly collected from Las Flores, Argentina where *S. richteri* is present, was established at the Mississippi sites since it preferentially attacks black and hybrid imported fire ants; at the other sites, *P. tricuspis*, a phorid that parasitizes red imported fire ants, has been released. Other potential differences in management of black, hybrid, and red imported fire ants are discussed.

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# **The Lago Santa Fe Fire Ant Project: An example of Community-wide imported fire ant management in Texas**

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Texas Cooperative Extension

## **Abstract**

Managing the red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae) has been demonstrated to dramatically reduce the cost insecticide use, maintain control of fire ants and eliminate problems caused by the ant. This demonstration, conducted in the Lago Santa Fe community in Galveston County, Texas, demonstrated several recent advances in conducting community-wide programs, including: 1) the effectiveness of the "hopper blend" treatment (50:50 hydramethylnon plus s-methoprene ant bait); 2) application methods such as the truck-mountable industrial "bait blower"; and 3) scheduling treatments to reach a goal of maximum control for an athletic event, the 2002 National Ski Championships.

## **Summary**

Managing fire ants on a community-wide basis at Lago Santa Fe, Santa Fe, TX has proven to be effective. Site assessment, proper application and judicious use of a hopper blend treatment of hydramethylnon + s-methoprene fire ant bait resulted in 89% control of fire ants in this community 8 weeks after application. As a result, there was minimal need of single mound treatments.

## **Problem**

The red imported fire ant, *Solenopsis invicta* (Buren (Hymenoptera: Formicidae), has become an important economic and health threat in urban Texas. According to a 1998 study conducted by the Department of Agricultural Economics, TX A&M University, of fire ant related costs in Dallas, Fort Worth, Austin, San Antonio, and Houston, fire ants have serious economic effects for these metro areas of Texas. Households experienced the largest costs among sectors examined with a average of \$151 per households spent annually which included repairs to property and equipment, first-aid, pesticides, baits, and professional services. Treatment costs accounted for over 50% of this total cost. A full damage assessment for Texas must include additional sectors, and the estimated costs of \$581 million per year for the selected sectors underscore the impact of this pest.

In Houston the average medical treatment costs per household of \$25.46. The duration of injury for children and adults was 6.6 days and 5.6 days, respectively. The fire ant limits outdoor activities and homeowners and producers incur added costs in managing the fire ant (see Lard et al. 2000, "The Economic Impact of the Red Imported Fire Ant on the Homescape").

## **Objectives**

During the past 5 years the Texas Imported Fire Ant Research and Management Project has championed the development of several "products/processes" that can be used in addressing the goal of eliminating the fire ant as a pest of major economic and health significance (Drees and Frisbie 2002). Fire ant management is centered around the "Two-Step Method" of fire ant control (see publications B-6043 and L-5070) which relies on the broadcast application of an effective ant bait



product, followed by selective individual ant mound treatment if necessary. One of the most publicly visible components of the project has been the demonstration of the concept of managing fire ants on a community-wide basis - even though funding has been restricted by the policy that no dollars could be used to buy or apply pesticides (i.e., no pesticide give-away programs).

Demonstrations of community-wide fire ant management have documented that the cost of pesticides people had been buying and applying to try controlling the ants on a property-by-property basis can be reduced by 84% (Riggs et al. 2002). Also, new tools to apply broadcast bait product treatments are in place and ready for use: 1) Descriptions for modifications needed to apply ant bait products by air are on the project's web site (<http://fireant.tamu.edu> under "management", entitled "Broadcast Application Guide"); 2) A prototype truck-mountable "bait blower" capable of treating neighborhoods by driving down the street at 10 to 30 miles per hour has been developed (Drees and Frisbie 2002). The mounting of bait spreaders to ATV's and other multiple use vehicles common to urban communities have been demonstrated and mounts for these various spreaders can be purchased through various dealers, and 3) Bait combinations and new "safer" bait active ingredients have been highlighted (see Fire Ant Trails 3(6), 5(2) and 6(2)).

Several recent advances in conducting community-wide fire ant management programs were highlighted in this demonstration, conducted in the residential community of Lago Santa Fe, Galveston County, TX. Lago Santa Fe is a private water ski community located 25 miles south of Houston, TX. This Community encompasses approximately 100 acres, with 4, 0.5 mile X 200 ft wide lakes, designed for water skiing events. Forty-eight homes border the 4 lakes in 1 acre lots. The objective was to utilize as many "items" championed by the Fire Ant Project in this community-wide effort.

## **Materials and Methods**

In the Fall of 2001, representatives from the Community of Lago Santa Fe asked the Texas Cooperative Extension for help in controlling fire ants in their neighborhood. This Private Lake Community was to host the 2002 National Ski Championships, and the U.S. Water Ski Open in August of 2002. They also were to host several regional events leading up to the 2002 Nationals and Open. They needed to control the fire ant since they were expecting around 5,000 visitors to their community for these upcoming events. This Community would also host these same events in 2003.

**Site and Fire Ant Activity Assessment.** Fire ant activity was determined by counting the active fire ant mounds in 0.25 acre circles in 8 undeveloped lots within the community (Table 1). A mound was considered active, if after disturbed with a prodding rod, fire ants emerged within a 15 second waiting period. An adjacent pasture area was monitored periodically to make sure fire ant activity monitored in the treated area was due to treatments and not other environmental conditions (Table 1).

**Products.** Because of the nearness of events (some starting in June) it was decided to use a combination of hydramethylnon and s-methoprene. The application of the 50:50 "hopper blend" of 0.75 lb. hydramethylnon fire ant bait (ProBait™, Amdro® or Siege® Pro Fire Ant Bait) blended with 0.75 lb. s-methoprene (Extinguish™ Professional Fire Ant Bait) or other "juvenoid" Insect Growth Regulator (IGR) fire ant bait product, applied 1.5 pounds of the blended products per acre has repeatedly resulted in a relatively quick and long-lasting suppression of red imported fire ant mound numbers in replicated tests (see <http://fireant.tamu.edu> under "research" and "applied research"). None of these products applied alone produces this effect.

This combination offered the quick action of metabolic inhibitor (hydramethylnon) to make sure fire ants were contained before the June events, and the extended activity of an IGA (s-methoprene), for



continued containment into the August event. Also, the Texas Department of Agriculture (November 30, 2001 memorandum from Phil Tham, Deputy Assistant Director for the Pesticide Programs Division) had issued a 24(c) (Special Local Needs) registration for the "hopper blend" application of Amdro® or Siege® Pro plus Extinguish Professional Ant Bait (SLN TX - 010016). We wanted to demonstrate that this combination would work well in an urban setting.

If extra baiting was necessary as the events dates drew closer, Justice Fire Ant Bait was chosen because of the active ingredient (spinosad) had been recognized as a "natural substance" by the National Organic Standards Board, and it is one of the fastest acting baits (see FAPFA039, "An Organic Two Step Method For Fire Ant Control" and Fire Ant Trails3(6) and 6(2)).

Finally, because we were trying to control fire ants around man made lakes that had several fish species, Orthene Fire Ant Killer containing 50% acephate was chosen over the synthetic pyrethroid dusts for single mound treatments to counter any run-off issues.

**Bait Applicators.** Three applicators/spreaders were used for the broadcasting of the fire ant bait. A handheld spreader set on the smallest setting was used. The homeowner or volunteer was given 1.5 lb of the blended bait product to spread over a 1.0 acre lot. A truck mounted "bait blower" (Drees and Frisbie, 2002) was used to spread bait along the roadside and in undeveloped lots. A GT-77 model Herd Spreader mounted to an ATV was used to spread bait around lake areas. Both the bait blower and ATV mounted spreader were calibrated to spread 1.5 lb bait/acre.

## **Results and Discussion**

Ant mound counts from 1/4 acre circles in 8 undeveloped lots (Table 1) showed an average of 168 mounds/acre. Baiting was initiated on April 18, 2002. Six weeks after treatments circle counts indicated control levels greater than 67%. A count taken in June before preliminary events were to start in July, showed control levels just above 89%.

A count taken one month before the 2002 Ski Nationals indicated control still better than 85%. It was decided at this time to go ahead and use the Justice Fire Ant Bait around those high-traffic areas where exhibitors would be placing product displays, to maintain a high level of control in that area. Heavy rains fell the day before the week long 2002 Ski National and 2002 Open events, and during the events. Greater than 9 inches of rain fell during that week.

Minimal use of Orthene was required to treat single mounds. Less than 1.5 lb of the Orthene Fire Ant Killer was used during this time, to treat mounds that appeared after all of the rain.

Well after the event, a final count taken before the scheduled fall applications of fire ant bait still showed greater than 60% control of the fire ant. It should be noted that fire ant pressure in the adjacent untreated pasture remained at the level for the duration of the study (Table 1). The Lago Santa Fe Community Fire Ant Project was great a success. During the event, informational packets about imported fire ant management were provided to participants together with temporary tattoos and T-shirts promoting the community-wide fire ant program. It showed that communities working together with the right tools used at the appropriate times will give good lasting control of a fire ant problem. The goal of 100% control may not be reached using the methods employed, but the chance that a resident or visitor may come in contact with the fire ant was dramatically decreased. Of all of the problems encountered during the 2002 Ski Nationals and 2002 Open, fire ants did not even make the list.



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## **Acknowledgments**

The author would like to thank Corrie P. Bowen for his assistance with the trial and Dr. Bart Drees for his help in the establishment of the trial and the statistical analysis of the data. He also appreciates Wellmark International and DowAgrosciences and Ortho for the generous donation of the fire ant control products used during this project.

Mention of a trademark or a proprietary product does not constitute an endorsement of the product by the Texas Agricultural Extension Service and does not imply its approval to the exclusion of other products that also may be suitable.

Extension programs serve people of all ages regardless of socioeconomic level, race, color, sex, religion, disability, or national origin. The TX A&M University System, U.S. Department of Agriculture, and the County Commissioners Courts of Texas Cooperating



**Table 1.** Red imported fire ant mounds per 0.25 acre circle plot, Lago Santa Fe, Galveston Co., Texas, treated with the hopper blend of Extinguish™ (s-methoprene) and ProBait™ (hydramethylnon) fire ant baits (0.75 lb each product blended together and applied using ground application equipment) on April 18, 2002.

Lot Number	Number of red imported fire ant mounds/0.25 acre		
	April 18 (pre-treatment)	May 28 (6 weeks)	June 12 (8 weeks)
<b>Treated area:</b>			
32	38	8	1
25	48	15	1
24	32	11	3
23	29	11	5
20	41	16	8
2	55	23	9
1	48	18	4
46	47	8	6
Mean $\pm$ Stand. Dev.	42.25 $\pm$ 8.88	13.75* $\pm$ 5.23	4.62* $\pm$ 2.97
<i>T</i> =		7.8243	11.3686
<i>n</i> = 8; d. f. = 8; <i>P</i> =		0.0000	0.0000
Percent reduction:		-67.46%	-89.07%
<b>Untreated area (plot):</b>			
1	34	27	28
2	27	28	17
3	12	10	13
4	14	15	17
Mean $\pm$ Stand. Dev.	21.75 $\pm$ 10.53	20.00** $\pm$ 8.91	18.75** $\pm$ 6.44
<i>T</i> =		0.2537	0.4859
<i>n</i> = 4; d. f. = 6; <i>P</i> =		0.4041	0.3221
Percent reduction:		-8.05%	-13.79%

\* Mean significantly different ( $P \leq 0.05$ ) from pre-treatment mean using the Student *T* test (Microstat).

\*\* No significant reduction in mean number of fire ant mounds per plot



abdomen is visible. Body remains of previously identified fire ants and native ants were used to compare and identify remains. The time used on cleaning up, sorting, and storing the samples was approximately 40 minutes and the time used on counting remains on each subsample was approximately 45 minutes.

## Results and Discussion

Fire ant segments (separate head, thorax or abdomen) predominated in *Dorymyrmex* refuse piles for both sample days. Whether *Dorymyrmex* ants preyed upon the fire ants or just collected them remains unclear. There were 6870 fire ant parts collected on July 31 and 2145 on August 8, they recovered 31% of fire ants parts in eight days being the heads the most frequent part found on the first sample day (28.47% ) and abdomens for the second sample day (7.40%) (Table 1). Pyramid ant nest where the complete midden was removed and no ant remains were left recovered a significant number of fire ants at a highly considerable rate during a short period of time (July 31 '02: AVG= 77.24; SE:18.15); August 8 '02: AVG=20.54; SE:6.11) (Figure 1). Body remains of five other ant species including pyramid ants were found on middens; *Dorymyrmex flavus*, *Pheidole* sp., *Monomorium minimum*, *Paratrechina* sp. and *Pogonomyrmex barbatus* but in extremely low numbers they don't represented more than 0.01% of the total samples, for both sample dates the highest recoveries were represented by fire ants in up to 99.9%. Somehow *Dorymyrmex* seems to be putting a lot of effort on "collecting" or "killing" fire ants and putting them on the refuse piles. If they are preying upon fire ants, this might be an indication of their role as potential biological control agent. Further studies are necessary to understand the interactions between *Dorymyrmex* and *S. invicta*, in particular case. when combined with bait applications. *Dorymyrmex* seems not to be affected by IGR bait treatments We speculate that *Dorymyrmex* in combination with IGR bait treatments may significantly extend the treatment interval required to maintain *S. invicta* densities below economic thresholds because *Dorymyrmex* interferes with *S. invicta* colonization and, even when established, interferes with *S. invicta* rate of increase to noxious densities, further and more detailed studies are needed.

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	Body	Head	Abdomen	Thorax	Head+Thorax	Thorax+Abdomen
July 31	7.61±1.38	<b>28.47±8.01</b>	22.72±6.77	12.13±3.11	1.49±0.41	4.83±0.95
August 8	1.50±0.49	4.9±1.72	<b>7.40±2.12</b>	4.68±2.16	1.41±1.18	0.64±0.21

Table 1. Mean and average of fire ant remains recovered from *Dorymyrmex flavus* middens



## Pastoral Peace?

### A third year report on *Thelohania solenopsae* in a Mississippi coastal pasture.

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## ABSTRACT

On October 19, 1999, *Thelohania solenopsae* (Microsporidia: Thelohaniidae) was introduced to nine polygyne red imported fire ant mounds in two plots located in a Mississippi coastal pasture. Ant samples were collected and mound number and population indices were recorded every few months for three years. Colony numbers and population indices for inoculated and control plots were not significantly different through the first two years even though second year values for inoculated plots were slightly lower than in the controls. Inoculated plots were, however, significantly lower in mound number ( $F=8.843$ ) and population index ( $F=14.235$ ) within the third year. *T. solenopsae* spore recovery occurred by the first sampling, 12 weeks post-treatment, and has continued at varying levels through the latest sampling. Highest numbers of spore positive colonies coincided with the third year difference in population between control and inoculated plots. Though this was a small field trial, significant reduction of imported fire ant populations and ability to sustain area infection over a three year span, demonstrates potential for use of *Thelohania solenopsae* as a biological control agent for suppression of imported fire ants.



## A Simulation Model of Competitive Interactions Among Polygyne Red Imported Fire Ant Colonies for Foraging Space and Food Resources

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A simulation model of polygyne (multiple queens) red imported fire ants, *Solenopsis invicta*, was developed. The foraging component integrated the foraging distributions of ants from several colonies in a population with predictions of colony level numerical dominance on simulated food baits. Foraging parameters included the distance to food resources, colony size (i.e. biomass), and average internidal (between nest) spacing among colonies. The colony with the highest potential of having the largest worker force available at a location was considered to be the numerically dominant colony at that location. Three analyses used independent performance statistics derived from observed data to test model performance.

The first analysis showed that the model did a significantly better job of explaining colony numerical dominance on baits than expected from a random assignment of colonies to be numerically dominant on simulated baits. The second analysis showed that the observed and expected frequency distributions of numerical dominance were significantly different when considering numerical dominance as a function of distance from colonies to baits. Food items within 200 cm could be correctly identified as to which colony would be the numerically dominant colony > 80% of time, and declined as the distance between colonies and baits increased. The third analysis showed that the observed and expected frequency distributions of numerical dominance were significantly different when considering numerical dominance as a function of observed ant abundance on baits. The model performed best when there were greater than 50 observed ants on baits.

The second component of the model, food harvesting, relies on parameters from the foraging component coupled with published data on daily energy requirements for colony maintenance and reproduction. The food-harvesting component uses predictions of the daily energy (joules per day) requirements for two food types and two physiological processes. Carbohydrates are required for colony maintenance and proteins for colony reproduction. The food-harvesting component is presented as a hypothetical scenario that dynamically changes each colonies predicted foraging area based on their food harvesting ability and colony demand for food resources. In the simulation, as colonies harvest food their foraging areas shrink in proportion to how much energy they have acquired in relation to an upper limit defined as their daily maximum energy requirement for reproduction or colony maintenance. Simulations depict changes in colony foraging areas as a result of food harvesting under different patterns of resource distributions. (e.g. regular, random, and clumped).



# Endocrine and Ovarian Changes in Newly Dealate Queens of

## *Solenopsis invicta*

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Studies were conducted on the physiological and hormonal changes following the release of alates from developmentally suppressive pheromones produced by mature queens of the fire ant *Solenopsis invicta* Buren. Winged virgin queens were removed from the pheromonal signal and placed in colony fragments. The time for dealation, degree of ovarian development, and biosynthesis rate and whole body content of juvenile hormone (JH) were measured. The production rate and content of JH were highly correlated. Dealation and the initiation of oviposition corresponded to peak production of JH. JH production rose sharply following separation from the natal nest, peaking after three days. After eight days of isolation, JH production gradually subsided to levels similar to that found in pre-release queens, but began to increase again after 12 days. Mature queens had highly elevated levels of JH relative to recently dealate females, probably reflecting the increased reproductive capability of these older females. The results support the hypothesis that the pheromone released by functional queens inhibits reproduction in virgin alates by suppressing corpora allata activity and the production of JH.



# Distribution Patterns of *Thelohania solenopsae* Spores in Red Imported Fire Ant Mounds in Southern Oklahoma

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## Abstract

*Thelohania solenopsae* (Microsporida: Thelohaniidae), a microsporidian, commonly parasitizes fire ants in Brazil and Argentina (Jouvenaz *et al.*, 1981). Recently, attempts have been made to use *T. solenopsae* as a potential biocontrol agent for Red Imported Fire Ant (RIFA), *Solenopsis invicta* (Hymenoptera: Formicidae). This study primarily focuses on detecting the presence of *T. solenopsae* spores in RIFA at three sites in Oklahoma, using two techniques, viz., Polymerase Chain Reaction (Snowden *et al.* 2001) and modified Trichrome staining (Kokoskin *et al.* 1999). In 1998, there were attempts to introduce *T. solenopsae* at two other sites in Bryan County, but the microsporidian spores never established. Similarly, another release in Carter County in 2000 also failed to establish. However, RIFA samples collected in October 2002 from two of our study sites, at Bryan Co., OK tested positive for this microsporidian, while the samples from the third study site at McCurtain Co. (120mi east of Bryan County sites) tested negative. The source of *T. solenopsae* spores in Oklahoma is unknown. Twelve out of fourteen and 17 out of 19 plots tested were positive for the *T. solenopsae* spores, in the two study sites at Bryan Co., and none of the 14 plots tested in the study site at McCurtain Co., had the spores. This study gains significance as this is the first confirmatory report of presence of this pathogen in RIFA workers, in Oklahoma. The results are promising and also widen the scope to study the distribution patterns of this protozoan in the populations of RIFA in Oklahoma.

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# **Influence of Phorid Flies and Low Humidity on Foraging Strategies of *Solenopsis invicta***

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## **Abstract**

The main objective of this research is to investigate the influence of phorids and low humidity on the foraging strategies of *S. invicta*. It is hypothesized that *S. invicta* foraging efficiency will be reduced under parasitoid pressure and low humidity common in a semi-arid desert environment. *Solenopsis invicta* colonies were collected near College Station, TX and Temple, TX. All colonies were polygyne, having multiple queens. The colonies were placed into an arena where foragers were exposed to phorid flies and low humidity. The colonies were starved for 48 hours then allowed to forage for 72 hours. Unlimited treatments were given a fresh three gram piece of hotdog every 24 hours. Food storage piles were located, dried, and weighed at the end of the experiment. *Solenopsis invicta* retrieved more food when food was unlimited regardless of phorid presence or humidity level. Food storage was similar across all treatments. Food was typically stored in the second level of the arena in low humidity and all phorid treatments. *S. invicta* may have overcome phorid pressure by using avoidance strategies.



# Seasonal Effects of Temperature on Red Imported Fire Ants (Hymenoptera: Formicidae)

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## Introduction

Temperature affects many aspects of red imported fire ant, *Solenopsis invicta* Buren, biology including metabolism, development rate, mating behavior, foraging activity, and colony maintenance. We have observed fire ants dying on disturbed mounds and on bare soil during hot, dry weather. An experiment was conducted to determine the effects of hot and cold conditions on fire ant mortality during different times of the year. Critical Thermal (CT) limits are temperatures at which the locomotor ability of the ant is so reduced that it can no longer escape conditions that would lead to its death. The CT limits, critical thermal maxima (CTMax) and critical thermal minima (CTMin), were determined for red imported fire ants in the laboratory over a 17 month period. Fire ants were collected monthly from a local field and tested within 6 hours of collection in a precision controlled computerized incubator affectionately named "Sputnik" (Fig.1).

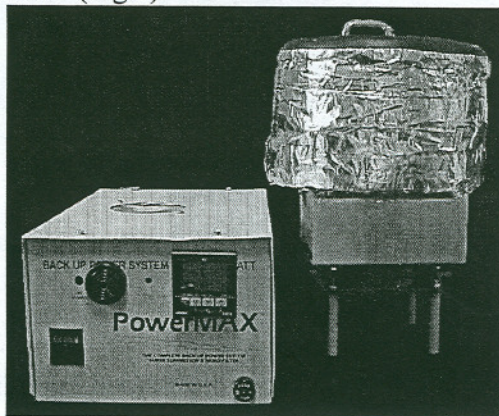


Fig. 1. "Sputnik" – a computerized incubator designed for CT determinations.

## Methods & Materials

Auburn's Research Instrumentation Group designed an apparatus specifically for CT determinations. "Sputnik" is a computer driven thermopile in which an electrical current produces the heat/cold. The computer can be programmed specifically for a task. For example, in our experiment, the computer is programmed to start at a designated temperature (ambient temperature) and raise or lower the temperature 1°C per minute to a destination temperature. A thermister near the bottom of the arena relays constant readings of the arena's interior temperature to the computer while a fan mounted to the Plexiglas® lid circulates air throughout the arena (Figs. 2 and 3).



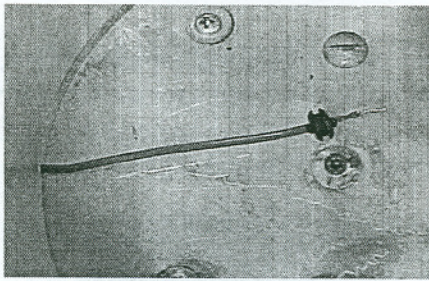


Fig. 2. Thermistor near bottom of arena

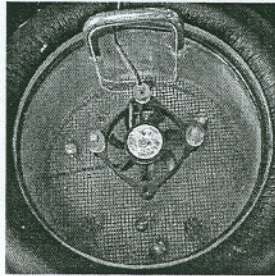


Fig. 3. Fan mounted on Plexiglas® lid

Red imported fire ant workers from monogyne colonies were collected the morning of the experiment and transported to the lab. Workers were individually weighed to the nearest 0.01 mg and placed individually into holding containers. Each holding container consisted of an open-ended glass tube painted with Fluon®. One end of the tube was covered with a piece of no-see-um netting, secured in place by a rubber band (Figs. 4 and 5).

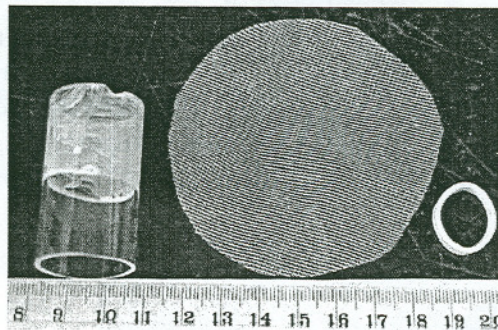


Fig. 4. Fire ant holding container components

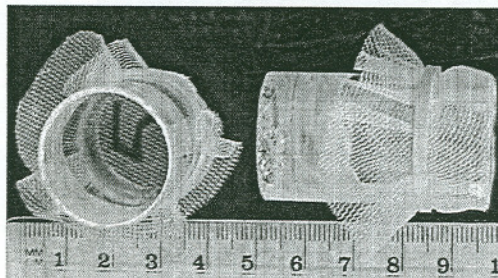


Fig. 5. Assembled fire ant holding containers



Ten numbered holding containers, each containing a single worker, were placed on a raised screen platform in the temperature controlled arena for each run along with a container of water to prevent desiccation (Fig. 6). The arena's temperature is raised or lowered from ambient temperature to determine the CTMax and CTMin, respectively.

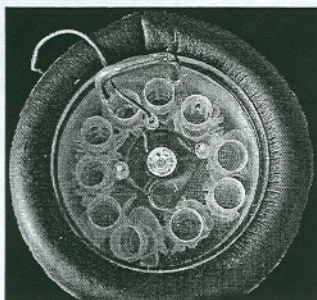


Fig. 6. Loaded arena with lid in place showing holding containers

## Results

The ants were observed until knockdown and the temperature recorded. Knockdown was defined as the temperature at which the ant became inactive or unable to move/right itself (Kay and Whitford 1978; Cockendolpher and Phillips 1990). Once knockdown was determined for a run, the ants were then removed from the arena and placed into a separate holding bin for recovery. 40 CTMax and 40 CTMin for a total of 80 measurements were taken each month for 17 months. Body mass measurements were taken for each ant. Both CTMax and CTMin were plotted against body mass (Figs. 7 and 8). Ant body mass was not related to either CTMax or CTMin at any test period (regression line not shown).

The data indicate that thermal changes in the red imported fire ant are a result of changes in environmental temperatures. Both CTMax and CTMin fluctuated with seasonal temperatures (Figs. 9, 10, and 11). However, a lag time appears to exist between the environment's temperature change and the ants' thermal change. These temperatures are varied, but CTMax's are generally greater than 30°C and CTMin's are 5°C and lower.



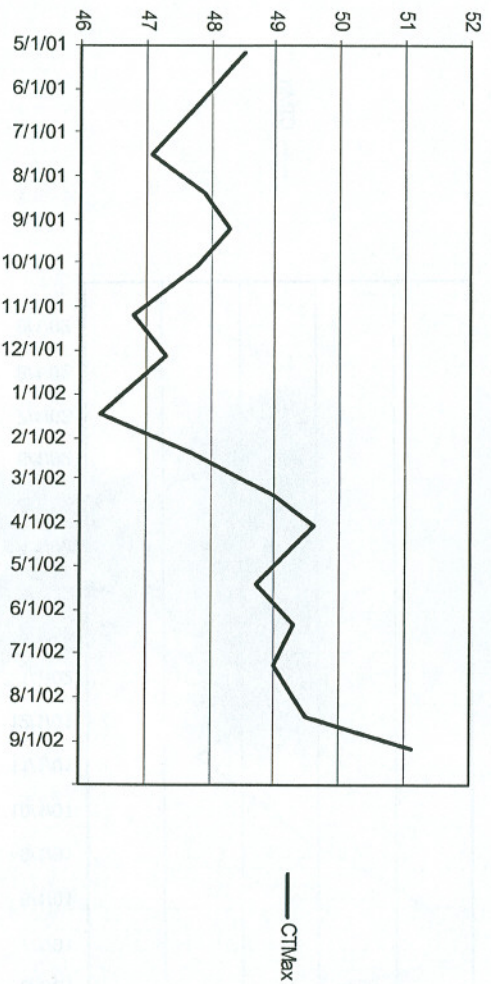
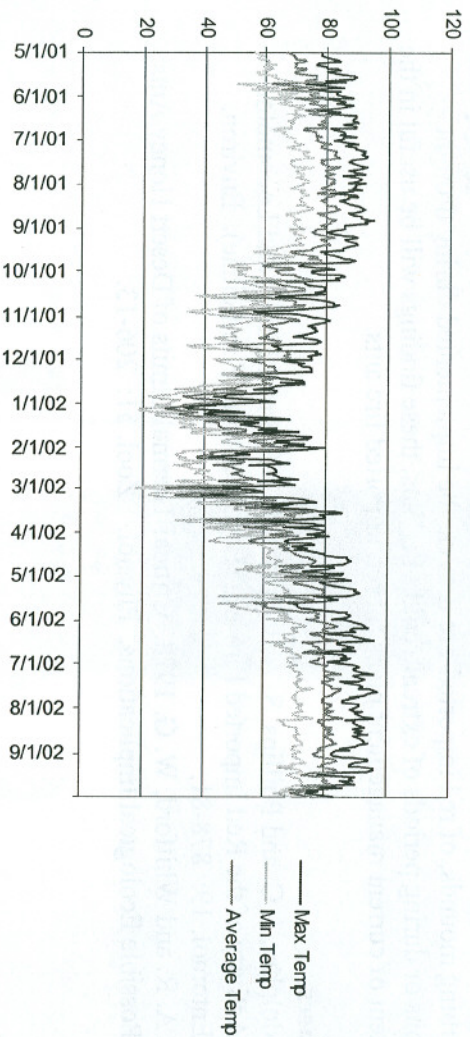


Fig. 9. CTMax temperatures

AWIS Max & Min





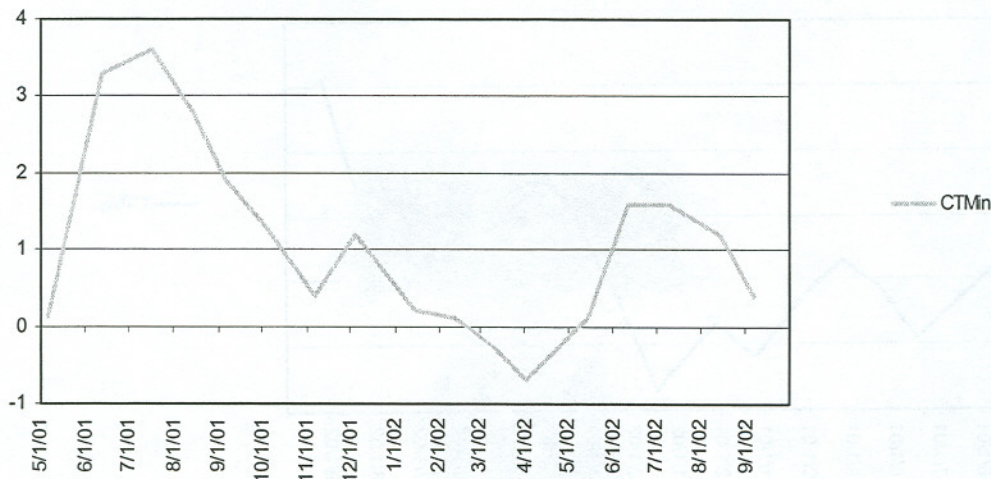


Fig. 11. CTMin temperatures

Ant temperature sensitivity varies with season and could be exploited during rapidly changing environmental conditions. For example, mechanical control, by dragging pastures or disturbing mounds, of red imported fire ants can be implemented during drought conditions or during periods of extreme cold. Perhaps, these findings will be useful in the refinement of current management plans for red imported fire ants.

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# Evaluation of Mechanical Disturbance of Mounds During Cold Weather on Red Imported Fire Ants

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## Introduction

Control of red imported fire ants, *Solenopsis invicta*, using baits or contact insecticides is cost prohibitive in many agricultural settings such as pastures. Shallow disking of a pasture or dragging heavy objects across the pasture are recommended nonchemical management strategies (Drees et. al., 2002). Mechanical control during mild weather conditions usually serves to temporarily flatten hardened mounds and some workers may be damaged. However, disturbing mounds during drought or cold weather may reduce populations by exposing the ants to adverse weather conditions.

The purpose of this study was to evaluate the effect of mechanical disturbance of mounds during cold weather on red imported fire ant populations.

## Materials and Methods

Three treatments were replicated four times in a randomized complete block design. The twelve plots were arranged in a 3.24 ha hay field. Plot size varied within the field to allow for approximately equal numbers of mounds within each plot.

Initial treatments were: 1) {cold} mound disturbance just before a cold front; 2) {warm} mound disturbance during average winter weather; and 3) an undisturbed control. A cold front was defined as a weather system predicted to have several nights below freezing with daily temperatures remaining below 10E C. Average weather was defined as weather with temperatures above 10E C and nights near freezing.

Plots were established and pretreatment population data were collected on October 23, 2001. Mounds were flagged, counted, and rated small (<100), medium (100-10,000) or large (>10,000) based on the number of workers present (Lofgren and Williams, 1982). A garden rake was used to simulate dragging in the cold and warm treatment plots.

Mounds in the warm treatment were disturbed on December 24, 2001 when the daily high was 20E C and the overnight low was -1E C. Mounds in the cold treatment were disturbed on December 26. The daily high was 7.8E C and the overnight low was -5.5E C. Night time temperatures remained at freezing or below until January 11, 2002.

Data were analyzed using the PROC GLM (SAS Institute, 1989b) and means were separated using LSD procedures.



## Results

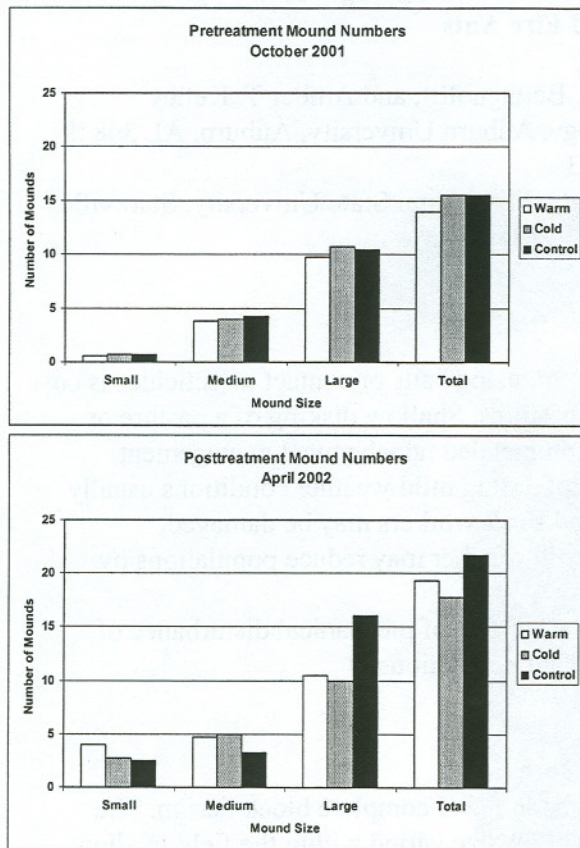


Fig. 1. In all treatments, total mound numbers were higher in April than in October. However, the number of large mounds decreased slightly in the cold treatment and increased in the undisturbed control.



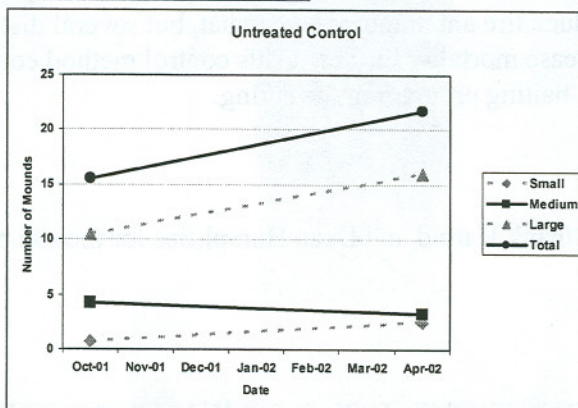
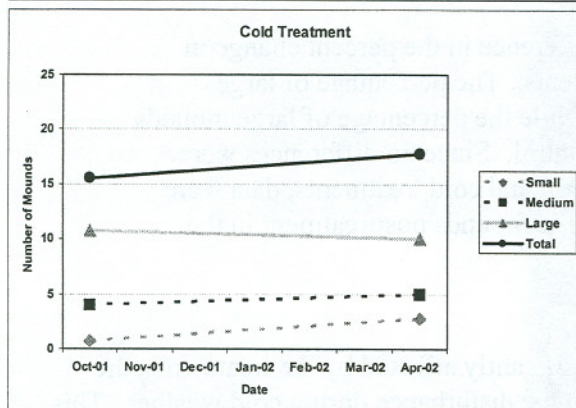
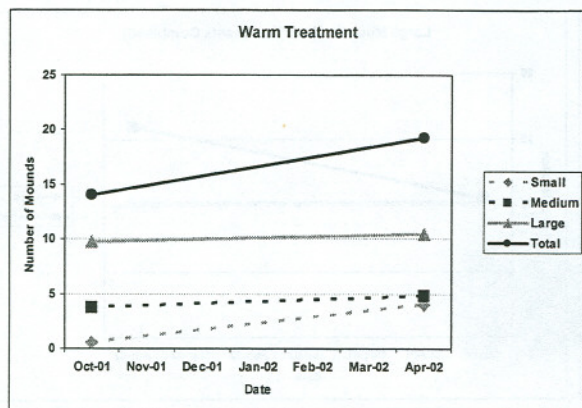


Fig. 2. Numbers of large mounds remained approximately the same pre- and posttreatment in both the warm and cold treatments while numbers of small and medium size mounds tended to increase slightly. This increase in small and medium mounds accounts for the increase in total mounds posttreatment in both treatments. In the untreated control, medium size mound numbers decreased slightly while numbers of large mounds increased, suggesting that the medium size mounds matured during the spring and developed into large mounds.



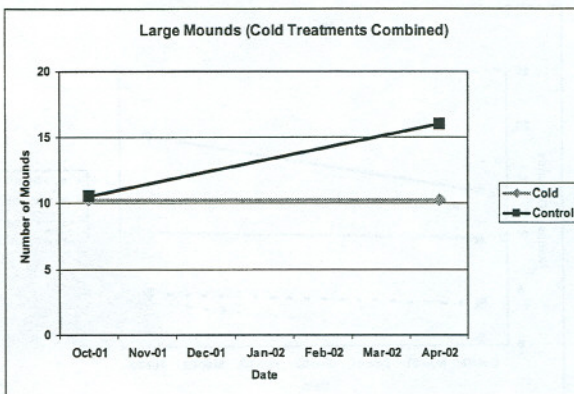
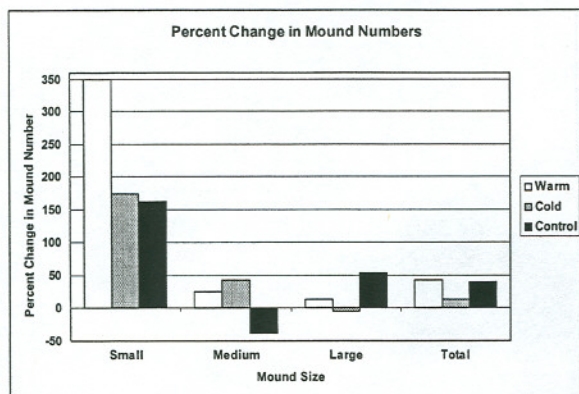


Fig. 3. There was a significant difference in the percent change in numbers of large mounds between treatments. The percentage of large mounds decreased in the cold treatment while the percentage of large mounds increased significantly in the untreated control. Since no differences were found between mound numbers in the warm and cold treatments, data were pooled. There were significantly more large mounds posttreatment in the control than in the disturbed treatments.

While total mound numbers were not significantly affected by the treatments, the number of large mounds decreased in response to the disturbance during cold weather. This method of control will reduce fire ant numbers somewhat, but several disturbances during the winter could possibly increase mortality further. This control method could improve the effectiveness of a planned baiting program in the spring.

## Acknowledgements

The authors would like to thank Harold and Dean Humphries for the use of their land and for help with the project.

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## Red Imported Fire Ants Reduce Lepidopteran Pests in Cotton but not in Soybean

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In previous studies we documented pervasive and often intense effects of predation by the red imported fire ant, *Solenopsis invicta* (Buren) (Hymenoptera: Formicidae), in cotton. Fire ants dramatically decreased the abundance of most arthropod herbivores, including lepidopteran larvae, on cotton plants both in greenhouse experiments and in the field. In this study, we describe greenhouse and field experiments that test for effects of red imported fire ants on lepidopteran larvae in soybean and we contrast these effects with the effects of fire ants in cotton. To quantify the relationship between red imported fire ant abundance and caterpillar abundance in cotton and soybean we sweep-sampled plots in both crops throughout the growing season in 1999. Fire ant abundance had a significant negative effect on lepidopteran larvae in cotton ( $R^2 = 0.27$ ) but not in soybean ( $R^2 = 0.006$ ). Using the field data, we conducted path analyses to quantify the direct and indirect interactions among fire ants, other natural enemies, and lepidopteran larvae. In cotton, the direct effect of fire ants on caterpillars was negative and fairly strong; however, fire ants also negatively affected the abundances of other natural enemies of caterpillars. Despite the indirect positive effects on caterpillar abundance, fire ants still maintained a negative net effect. In contrast, fire ants in soybean had a very weak direct effect on caterpillars, and strong but counteracting direct effects on two natural enemies; therefore fire ants had a very weak net effect on caterpillars. Finally, we conducted greenhouse experiments to quantify the survival of lepidopteran larvae in the presence and absence of red imported fire ants. Significantly fewer caterpillars survived in the presence of fire ants than in the absence of fire ants on cotton plants but there was no difference in caterpillar survival on soybean plants. These results are consistent with the field data that show that fire ants suppress lepidopteran pests in cotton but not in soybean. In cotton, fire ants tend cotton aphids (*Aphis gossypii*) and, as a result, spend more time foraging on foliage, thereby possibly accounting for stronger caterpillar suppression in cotton. Overall fire ant abundance (ground + canopy) was no greater in cotton than in soybean in 1999, but far fewer workers were sampled in soybean foliage. Recently established in the U.S., the soybean aphid (*Aphis glycines*) may provide us with the opportunity to test this idea as it expands its range south into the range of fire ants.



# Non-avoidance of Sodium Bicarbonate-treated Surfaces and Food by the Red Imported Fire Ant

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## INTRODUCTION

Sodium hydrogen carbonate ( $\text{NaHCO}_3$ ), also known as sodium bicarbonate, bicarbonate of soda, or baking soda is a registered pesticide active ingredient for use against fungal phytopathogens (EPA 1999). It controls powdery mildew (Horst et al. 1992) and inhibits growth of yeasts and bacteria (Corral et al. 1988). Effects of  $\text{NaHCO}_3$  on invertebrates are little known, but red imported fire ant, *Solenopsis invicta* Buren, mortality increased with increasing concentration of  $\text{NaHCO}_3$  in laboratory bioassays (Brinkman et al. unpublished data). The objective of this research was to determine whether fire ants are repelled by  $\text{NaHCO}_3$ . If  $\text{NaHCO}_3$  is not repellent to fire ants and causes relatively high levels of mortality, it may be useful as a safe and inexpensive control method.

## METHODS and MATERIALS

Fire ants were obtained from field populations in Spalding County, GA. These ants were removed from soil and maintained as laboratory colonies in plastic trays containing artificial nests. Ants were fed foods including 10% sugar water, tuna in oil, and yellow mealworm, *Tenebrio molitor* L., larvae.

**Arena Choice Tests.** A set of test arenas consisted of two 15.3 cm diam  $\times$  6.5 cm tall plastic dishes connected to a 4.5 cm diam  $\times$  4 cm tall Nalgene container with vinyl tubing. Fluon coated on inside walls of arenas prevented escape of ants. Artificial nests were placed inside each of the large plastic dishes. Sugar water was provided as a food source. Treatments of  $\text{NaHCO}_3$  were evenly spread on the bottom of nests and large containers at a rate of 18.0 mg per  $\text{cm}^2$ . Dental plaster in artificial nests was moistened with water prior to the start of tests; water was added as needed. In one set of arenas, large containers and nests on both sides received  $\text{NaHCO}_3$ . In another set of arenas, neither the containers nor nests were treated with  $\text{NaHCO}_3$ . In remaining arena sets, one side was treated by adding  $\text{NaHCO}_3$  to the large container and nest. All small containers were left untreated. Each test began by adding 100 workers to the small container. Mortality and number of live ants in large containers and artificial nests were recorded each day for 6 d. In the first test, there were five sets of arenas in which only one side was treated with  $\text{NaHCO}_3$  along with one set of arenas in which both sides were untreated (control), and one set of arenas in which both sides were treated with  $\text{NaHCO}_3$ . In subsequent tests, there were two sets of arenas in which either the left or right side was treated with  $\text{NaHCO}_3$ , along with two sets of arenas in which both sides were either untreated (control), or treated with  $\text{NaHCO}_3$ . These tests were conducted a total of three times between 11 June and 21 August 2002 using workers from three different colonies. Experimental design was a randomized complete block design (RCBD). Live ant data for artificial nests and large containers were analyzed separately and were also pooled for comparisons between treated and untreated sides of arena



sets. Data were analyzed by using the PROC MIXED procedure in SAS; significant means were separated with LSD ( $P = 0.05$ ).

**NaHCO<sub>3</sub>-Sugar Water Tests.** Test arenas were prepared by burning a 5-mm diam hole in the bottom of clear 35-ml plastic cups and adding dental plaster to about 10% of total cup volume. Fluon was applied to the inside walls of cups and undersides of lids. Ten workers were placed in each container. Cups were placed on a wet foam pad to maintain moisture in dental plaster within cups. Sugar water treatments were pipetted into 0.65-ml plastic containers. These were placed on the dental plaster in cups. Treatments that were tested were 10% sugar water (untreated control), 10% NaHCO<sub>3</sub> in sugar water, and 10% sugar water in a container placed next to a container with 10% NaHCO<sub>3</sub> in sugar water. Mortality was checked daily for 6 d. Treatments were replicated 10 times in a RCBD. These tests were conducted three times between 18 June and 04 August 2002 using workers from three different colonies. Data were analyzed by using the PROC MIXED procedure in SAS; significant means were separated with LSD ( $P = 0.05$ ).

## RESULTS

**Arena Choice Tests.** Although workers could avoid treated sides, they freely foraged in the large treated containers and also colonized treated nests. Mean number of live workers residing in treated sides (nest + large container) of arena sets did not differ significantly ( $F = 1.12$ ;  $df = 1, 7$ ;  $P = 0.3253$ ) from the number in untreated sides over the 6 days of the test (Fig. 1). The number of workers in both treated and untreated sides declined over time.

On three of the six sampling dates, the number of live ants in treated nests did not differ significantly ( $P > 0.05$ ) from the number in untreated nests. On the other sampling dates, fire ants were present in treated nests, but at lower numbers compared with untreated nests. There may have been some degree of preference for untreated nests, however the reduced presence of ants in treated nests was more likely due to mortality caused by treatments rather than potential repellency of NaHCO<sub>3</sub>. The mean number of workers in untreated nests on day 6 was less than half the number residing in those same nests on day 1.

Mean percent mortality of fire ants in arena sets in which neither side was treated was 25% on day 6 and was significantly ( $F = 22.44$ ;  $df = 2, 5$ ;  $P = 0.0001$ ) lower in comparison to arenas in which one or both sides were treated with NaHCO<sub>3</sub> (Fig 2). There was no significant ( $P > 0.05$ ) difference between mean percent mortality for arena sets in which both sides were treated with NaHCO<sub>3</sub> and mortality in arena sets with only one treated side.

**NaHCO<sub>3</sub>-Sugar Water Tests.** Cumulative mortality for fire ants provided untreated sugar water for 6d was significantly ( $F = 72.96$ ;  $df = 2, 5$ ;  $P = 0.0001$ ) lower than mortality for ants provided NaHCO<sub>3</sub>-sugar water mix and for ants that had a choice between feeding on NaHCO<sub>3</sub>-sugar water mix or untreated sugar water (Fig. 3). Cumulative mortality for fire ants provided sugar water mixed with NaHCO<sub>3</sub> did not differ significantly ( $P > 0.05$ ) from mortality for fire ants that could feed on NaHCO<sub>3</sub>-sugar water mix or untreated sugar water.



## DISCUSSION

The mode of action of  $\text{NaHCO}_3$  in fire ants is unclear. Efficacy of diatomaceous earth against insect pests has been attributed to the action of abrasion and desiccation (Carlson and Ball 1962) and some fire ants may have died from those stressors after foraging in  $\text{NaHCO}_3$ -treated containers. However, Brinkman and Gardner (2001) exposed fire ant workers to diatomaceous earth and observed much lower mortality than was observed for fire ants exposed to  $\text{NaHCO}_3$  in this study. Corral et al. (1988) and Horst et al. (1992) attributed the antimicrobial activity of  $\text{NaHCO}_3$  to bicarbonate ions and elevation in pH. Ants frequently clean appendages and this behavior would have facilitated *per os* entry of  $\text{NaHCO}_3$ . Sodium hydrogen carbonate contributes  $\text{OH}^-$  to solution and large amounts would increase internal pH (Tortora and Grabowski 1996). An increase in internal pH caused by ingestion of  $\text{NaHCO}_3$  may have interfered with enzymatic activity in fire ants and caused their deaths. The optimal pH range for enzymes is narrow (Chapman 1982), and departure from narrow limits of normal  $\text{H}^+$  and  $\text{OH}^-$  concentrations disrupts enzymatic functions (Tortora and Grabowski 1996).

Direct observations of ant feeding were not done, but mortality for ants provided a choice between sugar water and  $\text{NaHCO}_3$ -sugar water mix was higher than for ants provided sugar water only. These results suggest that ants were not repelled by  $\text{NaHCO}_3$  in sugar water and were killed following ingestion. This mortality was relatively moderate (=50%). It is not known if higher levels could be achieved by using higher concentrations of  $\text{NaHCO}_3$  in sugar water or another carrier. If  $\text{NaHCO}_3$  can be distributed by trophallaxis throughout an ant colony, it may be useful as an active ingredient in an ant baiting control strategy.

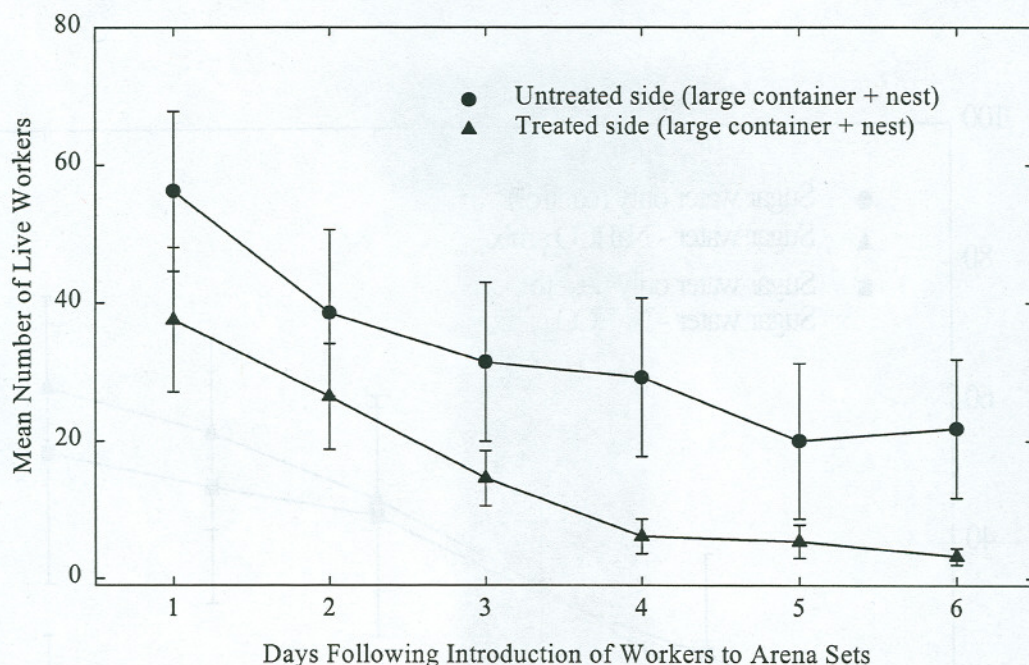
Sodium hydrogen carbonate or baking soda is safe for humans, inexpensive, and potential exists for use in fire ant IPM. Additional studies should be conducted to determine if  $\text{NaHCO}_3$  can be used in baits and whether it can protect structures such as utility boxes from colonization by fire ants.

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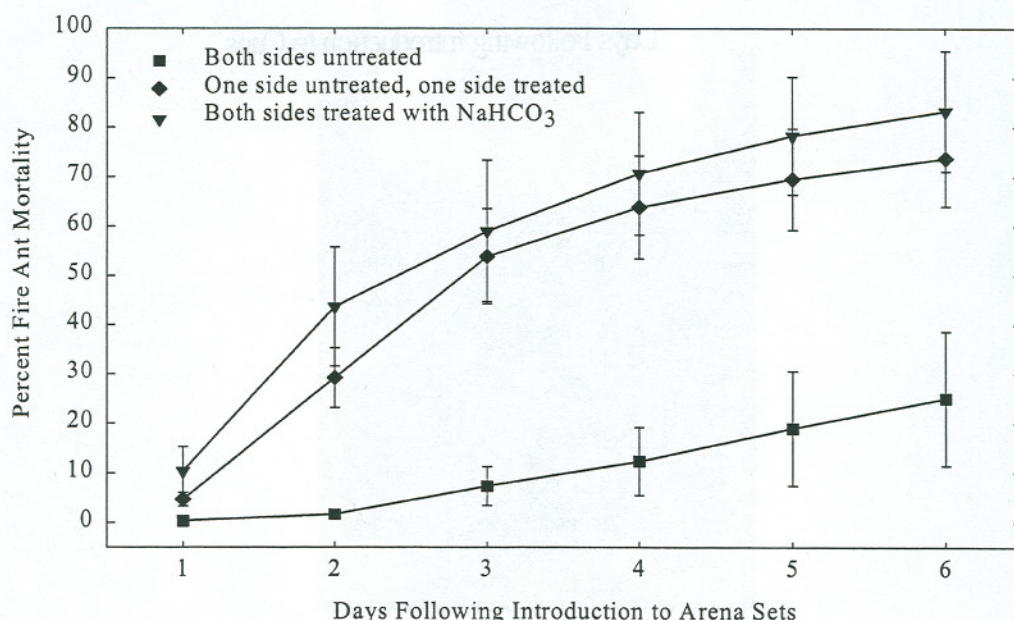
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**Fig. 1.** Mean number of live *S. invicta* in untreated sides of arena sets and in sides treated with  $\text{NaHCO}_3$ . Ants ( $n = 100$ ) were added to a smaller central container and could move freely between treated and untreated sides through vinyl tubing. Each side was comprised of a large container plus artificial nest.

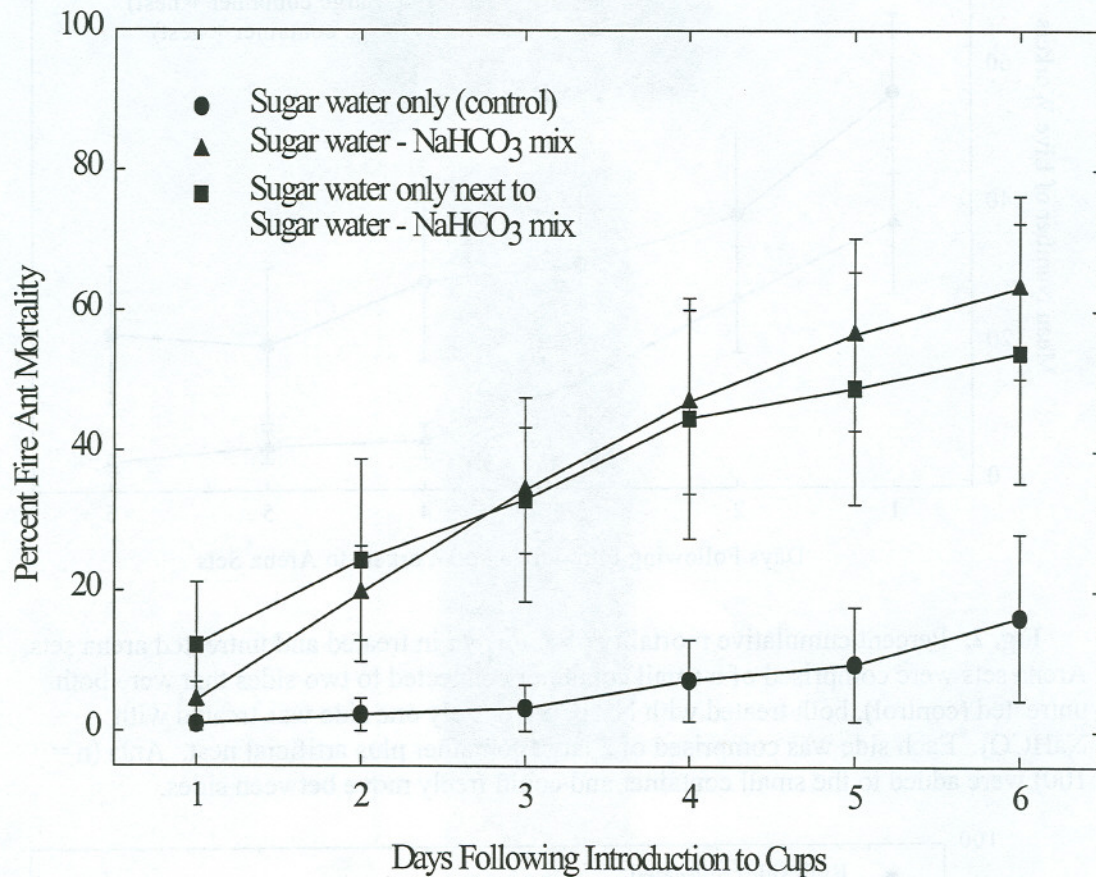


**Fig. 2.** Percent cumulative mortality of *S. invicta* in treated and untreated arena sets. Arena sets were comprised of a small container connected to two sides that were both untreated (control), both treated with  $\text{NaHCO}_3$ , or only one side was treated with  $\text{NaHCO}_3$ . Each side was comprised of a large container plus artificial nest. Ants ( $n = 100$ ) were added to the small container and could freely move between sides.





**Fig. 3.** Percent cumulative mortality of *S. invicta* provided untreated sugar water (control), NaHCO<sub>3</sub>-sugar water mix, or untreated sugar water in a container next to a container with NaHCO<sub>3</sub>-sugar water mix.





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*(Note: These “snapshots” are single page printouts of each poster. The original Poster files are on the CD provided with these Proceedings and are identified by the page numbers and first author, as listed below.)*

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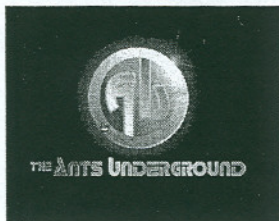
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# The Ants Underground: Youth CD-ROM and Lesson Plans

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Chris Meux<sup>4</sup>, Jason Shivers<sup>4</sup>, and Ed Rhodes<sup>4</sup>, J. Brian Richardson<sup>5</sup>

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Ants Underground Logo

## Abstract

Youth education is important and is a part of the fire ant education program. An interactive project aimed at educating youth aged 9 - 12 has been developed. Fire ant history, biology, and management is presented to youth using animation and sound.



Depiction of Red Imported Fire Ants

## Introduction

Our youth fire ant education program has three major objectives:

- For youth to develop an understanding of basic fire ant biology
- For youth to develop an understanding of proper management techniques, and the consequences of use of improper methods
- For adults to be impacted by youth, i.e., teaching youth may alter behavior of parents, especially in the common use of gasoline (Blanchard, 2000)

While one-time visits to a classroom are successful in introducing the youth to fire ants, a more in-depth presentation of fire ant information was needed to have an impact and change youth's attitude toward fire ants. Experience in the classroom with other educational materials (Project Wet, Project Wild) has shown that reinforcement of ideas increases the level of learning. Therefore, in June 1997 a Cooperative Extension Service workshop discussed the development of the youth program using computer technology

## Production and Evaluation Stage

Fire ant education was the purpose of the youth cd-rom. However, we wanted the program to be interactive. Interaction to include selection of content view and opportunities to answer questions via computer. Areas of emphasis were to be the history, biology, and management of the red imported fire ant.

The storyboards were developed (Fig 1) and clay models (Fig 2). Lightwave (Newtek) and Poser (Curious Labs) were the 3D animation tools. Flash 4 was used for authoring, programming and interface design. Flash 4 and AfterEffects (Adobe) were used for 2D animation. Evaluation among Extension personnel has been a part of the program since its inception. Children of CES personnel have been asked to critique the material, and Pulaski Co. 4-H groups have been involved on a formal basis. The initial evaluation by Arkansas 4-Hers helped us in identifying problem language areas and the suitability of graphics. An evaluation of the games segment was conducted in Fall 2000 and Winter 2003. McPeake & Ballard (2000) videotaped the youth volunteers playing the games in Fall 2002, and a written evaluation was prepared. Based on the results of 2000 and 2003 there were several suggestions for improvement in the games section of the program.

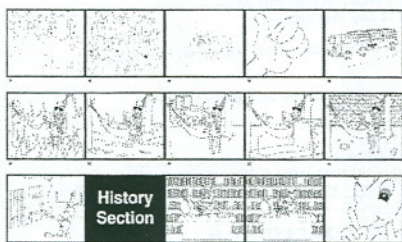


Fig 1. Storyboard



Fig 2. Clay model of ant figure

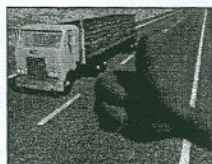


Fig 3. Image from General Debriefing

## Production Completed

"The Ants Underground" provides an alternative to the traditional teaching methods that may reach a child not responsive to traditional teaching methods. Hart (1983) cited that a wide range of teaching techniques directly enhances the learning process and we feel "The Ants Underground", provides a wide-range of possibilities for its use. "The Ants Underground" story is told from the perspective of a native ant in the general debriefing section, and continues to the history, biology, and management. The program concludes with the participant receiving a clearance badge to go out develop a plan of action for their community.

We have developed teacher lesson plans that tie into the current scientific education standards as part of "The Ants Underground" package. This tie-in is critical to reach a larger audience with our educational effort, by assuring the project's use by educators.

We have been evaluating the project from the storyboard stage and now are ready for its true field evaluation with teachers and other educators. After several revisions "The Ants Underground" is completed and will be distributed throughout Arkansas. We are in the process of developing training workshops for agents and teachers to assist in the successful use of the program.

Development of "The Ants Underground" has been very challenging. The rapidly changing technology has impacted the completion deadline. Use of the technology, and the financial resources involved in the production of the project were major concerns for the project.

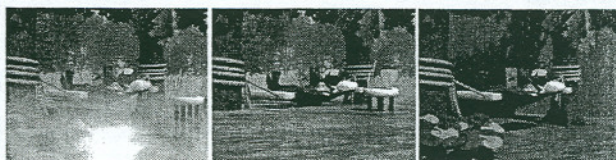


Fig 4. Sequence of images from General Debriefing

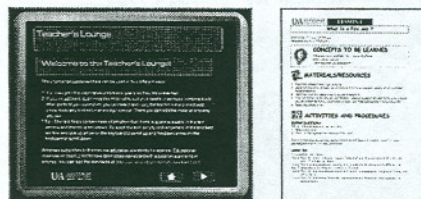


Fig 5. Teacher Lounge Page and PDF version of Lesson 1

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# Cindy Heights, Neighborhood Abatement Program, Jefferson County, Arkansas

Joe Ivy<sup>1</sup>, Donna Shanklin<sup>2</sup>, Kelly Loftin<sup>3</sup> and John Hopkins<sup>3</sup>

University of Arkansas Cooperative Extension Service



## Introduction

Neighborhood abatement programs are difficult to initiate in Arkansas. Communities have to take ownership in the program for the program to continue. In 2002, an African-American neighborhood began the first step of developing a program. Gaining confidence in the Extension-recommended Two-Step method of fire ant management was the first step. Demonstrating the program's effectiveness and the neighborhood's own ability to minimize the presence of the red imported fire ant by community involvement was the second step.

The red imported fire ant is well established in Jefferson County, the county having been under the USDA/APHIS Quarantine since 1990. Pine Bluff with a population of 55,085 is the largest community in Jefferson Co. (population 84,278). The Cindy Heights Subdivision is located in the central area of the city, surrounded by schools and other subdivisions (Fig 4). It is not an isolated area.

## Materials and Methods

Community fire ant abatement programs are a part of many communities in Arkansas. Unlike demonstrations or educational programs where state Extension faculty or county Extension agents are responsible for program implementation, the community itself takes ownership of the program with assistance from Extension. Members of the Cindy Heights Neighborhood initiated a request to the Jefferson Co. Extension office for an abatement program in the summer 2002. Extension personnel became involved in trying to assist them with this request.

Fire ants were managed using the Extension-recommended Two-Step method. In late summer August 2002 the 68 house neighborhood was treated by broadcasting a bait product throughout the neighborhood (Fig 1, and Fig 2) A contact product was provided to street captains to treat individual mounds on an on-request basis.

A house-to-house survey was conducted December 5, 2002 to assess individuals in the neighborhood's opinion about the program. Our goal was to assess program participation and effectiveness in changing perceptions about fire ant abatement and control methods. These face-to-face interviews were conducted during daylight hours by visiting the respondents at home (Fig 3). Home addresses of respondents were mapped (Fig 4).

The interviewers worked together and questioned adult household members > 17 years old. After a brief introduction explaining the interviewers' affiliation and purpose of the survey, one interviewer asked and another recorded responses to five questions. Questions were used to elicit a wide-range of responses about the program, its successes, and its failures. These questions were: 1: What did you think of the treatment program? 2: Did you think it had an impact? 3: What were you using prior to the treatment? 4: Would you be willing to continue the program and if so. 5: Any comments you would like to make

## Results and Discussion

One-on-one personal interviews were conducted by three interviewers on December 5 2002. Nineteen homes (28%) of the 67 homes in the subdivision were involved in the survey. Nineteen interviews were initiated, however only 17 were completed. Responses were recorded on paper and compiled. The average interview length was less than 5 minutes.

In this small neighborhood, the interviewers and respondents were familiar with one another prior to the interview. One interviewer was a street captain and had been involved in the abatement program from its initiation, the other interviewers were the local county extension agent and an extension specialist. There was no need to recruit a minority interviewer in that two of the three interviewers were African-American males. It is certain that their presence increased the response rates and openness of minority respondents (Tashakkori & Teddlie 1998). Studies show that the race of an interviewer can play a role in response bias, particularly if a standardized measurement process is not followed (Fowler & Mangione 1990). Weiss (1968) found in a study of welfare mothers that a higher rapport with the interviewer resulted in biased data, and this may have brought a certain bias to the survey. Because interviewers were known by respondents prior to the interviews, the interviewers stressed the need to know the good and bad about the program throughout the interview (Tashakkori & Teddlie 1998). A visual inspection indicated all block areas in the abatement program were sampled (Figure 4). The sample was deemed satisfactory. There were 9 male respondents and 8 female.

The following is a general overview of responses:

**What did you think of the treatment program?**

Good, Fine - 15  
Neutral - 2  
Negative - 0

**Did you see any changes in fire ant activity?**

Yes - 15  
No - 0  
Do not know - 2

**What were you doing to manage your fire ants previously?**

Nothing - 2  
Unknown contact insecticide - 10  
Pest Control Operator - 2  
Gasoline - 1  
Hot Water - 1  
Amdro - 1

**Do you think the neighborhood will continue the program? Why or Why Not?**

Yes, I would like to see it continue - 16  
Check with my husband - 1

**Would you be willing to pay for the treatment? If so, How Much?**

Captain Johnny Jones asked if it was \$20.00 for a spring and fall treatment would they be willing to pay that amount.

Yes - 17  
Typical replies (I would be willing, I can afford, and you are spending that much yourself)

**Any Comments about the program you would like to make?**

Glad to see the Ag program at work  
Could we extend it to surrounding areas  
I think it was nice the ants left  
I didn't know what was going on, but noticed a decrease in fire ants  
Glad to see someone take an interest in this neighborhood  
That's good. It needs it - suckers were getting out of hand

## CONCLUSION

Review of the responses show that the majority of the residents were satisfied with the results of the treatment. However, many were still unaware of the two-step program. From the terminology used by the respondents many have not taken ownership in the program in that they consistently stated 'I hope THEY will continue the program'. Further meetings are planned for 2003 to educate the neighborhood more extensively in the two-step fire ant management program and what their neighborhood can do to minimize fire ants.

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Fig 1. Street Captains and CES agent Ivy Prepare to treat neighborhood



Fig 2. CES Agent Ivy Evaluates effectiveness

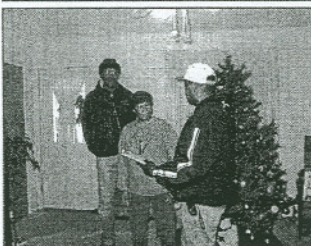


Fig 3. Street Captain Johnny Jones and CES agent Ivy questioning Emma Jones, neighborhood member

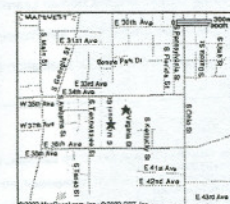


Fig 4. Cindy Heights Neighborhood



# Evaluation of broadcast applications of various contact insecticides

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## Abstract

The objective of this trial was to evaluate the efficacy of broadcast applications of 0.0103% fipronil granules, 0.1% cyfluthrin granules, 0.2% imidacloprid granules, and two rates of cyfluthrin/imidacloprid liquid against red imported fire ants, *Solenopsis invicta*. At one day post-treatment the imidacloprid granular, cyfluthrin/imidacloprid 3 oz/acre liquid, and cyfluthrin granular treatments had statistically significant fewer fire ants than the untreated control. The estimated percent reduction was 90, 66, and 45 for imidacloprid granular, cyfluthrin granular and cyfluthrin/imidacloprid 3 oz/acre liquid, at day 1 post treatment, respectively. The imidacloprid granular treatment maintained control through 7 days. The bifenthrin and fipronil granular treatments showed significant reduction at 3 days. Fipronil treatment was the only insecticide treatment to show a statistically significant reduction in fire ants and the proportion of mounds containing brood (egg, larva, pupa). This amounted to a 56% reduction when compared to the untreated control.

## Materials and Methods

**Study Site:** The study was conducted in Rye, Arkansas located in Cleveland County in southeast Arkansas. Density of red imported fire ant, *Solenopsis invicta* mounds at the study site was approximately 240 mounds per acre. Grass had been mowed approximately 3 days prior to the study.

**Design:** Each treatment and the untreated control was replicated in 3 plots. Plots were approximately 0.25 acres in size. The treatments and control were randomly allocated within each block. The three blocks corresponded to different areas of the study site.

**Evaluation:** Pre and post treatment evaluations were conducted using bait stations to collect foraging ants within each plot. Bait stations consisted of a 1/4 inch hot dog cube placed on a snap vial lid and marked with a wire survey flag. Ten bait stations (two transects with 5 bait stations each, located in the center of each plot, transects were approximately 30 feet apart, bait stations within each transect were approximately 30 feet apart) in each plot were made available to foraging ants for approximately 30 minutes. The numbers of red imported fire ants estimated at each station in each plot was used to evaluate product efficacy. Percentage of mounds containing brood was determined during the final evaluation. Ten mounds in each plot were excavated and observed for the presence of fire ant brood (eggs, larva and pupa).

**Insecticide Applications:** Products and application rates are given in Table 1. Insecticides were broadcast using a Herd® seeder mounted on a Kawasaki® mule equipped with a digital speedometer. After calibration, the specified rates were broadcast by matching to the appropriate ground speed and dispersal rate. Liquid insecticide applications were made with a trailer mounted boom sprayer pulled by a Kawasaki® mule equipped with a digital speedometer. After calibration of the boom sprayer, a test application outside of treated area and with water only was made to determine the speed and amount of water required for uniform coverage. Insecticide applications were made on August 6, 2002.

**Statistical Analysis:** Data collected from bait stations were analyzed using Analysis of Variance based on a RCB design with 3 replicates. The protected LSD procedure was used to determine significant differences in the mean number of foraging RIFA=s collected from the various treatments (Statistix 2000). The percentage of mounds containing brood was analyzed using the same procedures as above.

Table 1. Insecticide rates and manufacturer

Insecticide	Rate	Manufacturer
0.25% permethrin GR	87lb/acre	Real Kill Multi Purpose
0.147% bifenthrin GR	136lb/acre	Scotts MaxGuard Insect Protection with Turf Builder
0.2% imidacloprid GR	65lb/acre	Bayer Advanced Lawn Grub Control
0.0103% fipronil GR	87 lb/acre	Over N Out
0.1% deltamethrin GR	87 lb/acre	Delta Shield
0.72% cyfluthrin EW and 0.72% imidacloprid	43.5oz/acre	Research stock supplied by Bayer-Purcell LLC.
0.72% cyfluthrin EW and 0.72% imidacloprid	130.7oz/acre	Research stock supplied by Bayer-Purcell LLC.
0.1% cyfluthrin GR	130lb/acre	Bayer Advanced Power Force



## Results and Discussion

The mean number of foraging fire ants estimated at bait stations is given in Table 2. At one day post-treatment the imidacloprid granular, cyfluthrin/imidacloprid 3 oz/acre liquid, and cyfluthrin granular treatments had statistically significant fewer fire ants than the untreated control. The estimated percent reduction was 90, 66, and 45 for imidacloprid granular, cyfluthrin granular and cyfluthrin/imidacloprid 3 oz/acre liquid, at day 1 post treatment, respectively (Table 3). The imidacloprid granular treatment maintained this trend through 7 days post treatment. The bifenthrin and fipronil granular treatments began showing significant reduction in the number of ants collected from bait stations at 3 days post treatment. The fipronil granular treatment maintained significant levels of control only through 14 days post treatment. Significant reductions in the number of ants observed at bait stations were never achieved by deltamethrin, permethrin or the cyfluthrin/imidacloprid 1 oz/acre combination. By 65 days post treatment only the fipronil plots had statistically significantly fewer fire ants than the control.

The mean percentage of fire ant mounds with brood is given in Table 2. Fipronil treatment was the only insecticide treatment to show a statistically significant reduction in the proportion of mounds containing brood (egg, larva, pupa). This amounted to a 56% reduction when compared to the untreated control (Table 3).

Table 2. Mean number of red imported fire ants per bait station. Brood (far right column) is the percentage of fire ant mounds containing brood (eggs, larva, pupa).

Insecticide	DAYS							
	0	1	2	3	7	14	35	65
Untreated Control	32.5	50.8 <sup>a</sup>	36.5 <sup>a</sup>	22.2 <sup>ab</sup>	71.7 <sup>ab</sup>	42.8 <sup>ab</sup>	55.8 <sup>ab</sup>	56.8 <sup>ab</sup>
0.25 % Permethrin granule	52.0	52.5 <sup>a</sup>	38.3 <sup>a</sup>	33.5 <sup>a</sup>	70.0 <sup>ab</sup>	50.0 <sup>ab</sup>	57.5 <sup>a</sup>	70.8 <sup>a</sup>
0.147% bifenthrin granule	34.0	34.2 <sup>abc</sup>	30.0 <sup>a</sup>	15.8 <sup>abc</sup>	34.6 <sup>ab</sup>	18.7 <sup>cd</sup>	42.0 <sup>ab</sup>	37.0 <sup>ab</sup>
0.2% imidacloprid granule	49.2	5.2 <sup>d</sup>	7.5 <sup>b</sup>	4.2 <sup>d</sup>	19.8 <sup>a</sup>	37.7 <sup>abc</sup>	36.7 <sup>b</sup>	58.5 <sup>ab</sup>
0.0103 fipronil granule	37.5	49.2 <sup>ab</sup>	27.8 <sup>a</sup>	9.5 <sup>cd</sup>	43.3 <sup>abc</sup>	13.5 <sup>d</sup>	9.50 <sup>d</sup>	16.0 <sup>d</sup>
0.1% deltamethrin granule	39.5	39.7 <sup>abc</sup>	31.1 <sup>a</sup>	20.8 <sup>ab</sup>	55.0 <sup>abc</sup>	35.0 <sup>abc</sup>	45.8 <sup>ab</sup>	61.2 <sup>ab</sup>
0.72% cyfluthrin + 0.72% imidacloprid spray (1.0oz/100 sq ft)	33.3	40.4 <sup>ab</sup>	34.2 <sup>a</sup>	23.3 <sup>ab</sup>	70.0 <sup>ab</sup>	54.2 <sup>a</sup>	48.3 <sup>ab</sup>	61.2 <sup>ab</sup>
0.72% cyfluthrin + 0.72% imidacloprid spray (3.0oz/100 sq ft)	40.8	27.8 <sup>cd</sup>	32.3 <sup>a</sup>	21.7 <sup>ab</sup>	75.8 <sup>a</sup>	52.0 <sup>a</sup>	60.1 <sup>a</sup>	60.0 <sup>ab</sup>
0.1% cyfluthrin granule	31.0	17.0 <sup>cd</sup>	21.3 <sup>ab</sup>	21.5 <sup>ab</sup>	50 <sup>abc</sup>	32.2 <sup>cd</sup>	46.0 <sup>ab</sup>	53.8 <sup>ab</sup>

Means followed by the same letter are not significantly different using the protected LSD mean separation procedure ( $\alpha = 0.05$ ).

Table 3. Estimated percent control of red imported fire ants after broadcast application with selected insecticides. Brood represents the estimated percent reduction in the proportion of mounds containing brood (eggs, larva, pupa).

Insecticide	DAYS							
	1	2	3	7	14	35	65	Brood
0.25 % permethrin granule	0	0	0	2	0	0	0	0
0.147% bifenthrin granule	33	18	29	51	56	25	33	5
0.2% imidacloprid granule	90	79	81	72	12	34	0	11
0.0103% fipronil granule	3	24	57	40	68	83	72	56
0.1% deltamethrin granule	22	15	6	23	18	18	0	0
0.72% cyfluthrin + 0.72% imidacloprid spray (1.0oz / 100 sq ft)	20	6	0	2	0	13	0	5
0.72% cyfluthrin + 0.72% imidacloprid spray (3.0oz/100 sq ft)	45	11	2	0	0	0	0	0
0.1% cyfluthrin granule	66	42	3	30	27	18	5	5

Means followed by the same letter are not significantly different using the protected LSD mean separation procedure ( $\alpha = 0.05$ ).

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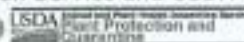
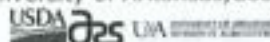
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# Biological Control Releases in Arkansas 2002: *Pseudacteon tricuspis* and *Theiohania solenopsae*

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## Abstract

The phorid fly, *Pseudacteon tricuspis*, and the microsporidian *Theiohania solenopsae* were released in Arkansas in 2002. Approximately 2000 and 1200 phorid flies were released in Pike and Bradley Counties, respectively. Initial results from Pike Co. suggest that the phorids have reproduced and expanded out of the immediate area of the release. Twenty-five fly and mound in Miller Co. were inoculated with *Theiohania solenopsae* infected brood.



Fig. 1. 2002 Release Sites in Arkansas  
(*Pseudacteon tricuspis*)



Fig. 3. Phorid feeding



Fig. 3. *Theiohania* spores



Fig. 4. *Theiohania* spores in mound

## Introduction

Classical biological control attempts to release predators or parasites with their prey or host. Arkansas is attempting to release the phorid fly *Pseudacteon tricuspis*, and the microsporidian *Theiohania solenopsae* with its natural host the red imported fire ant *Solenopsis invicta* (Fig 1).

*Pseudacteon tricuspis* is a parasitoid fly. The egg is deposited by the female into the head and pupal development occurs in the head of the fly. In the process of developing an enzyme is released by the fly which causes the tissues of the thorax to liquefy and the head falls off (Fig 2). This makes the fly a "reflexive" biological control organism, it is the behavior of the ants which when the flies are present - they hide, and are not efficient foragers (Fig 22).

*Theiohania solenopsae* is the most common fire ant pathogen in Brazil. It was discovered in the US in 1998 (TX, MS, OK). The microsporidian is an obligate intracellular parasite which impacts the longevity of the ant (Fig 3 and Fig 4). The result is decreased colony size and colony density.

## *Pseudacteon tricuspis* Release Method

Using protocols developed at USDA-ARS Gainesville, Florida the phorid fly, *Pseudacteon tricuspis*, was released in Pike and Bradley Counties, Arkansas.

The sites were evaluated prior to the release for the presence of various habitat characteristics. Characteristics including a high population of fire ants - preferably monogyne colonies, the topography of the area suggested should be diverse - including changes in elevation, diverse plant material from weeds to trees, and a water source. (Fig 5 and Fig 6). Both the release and control sites met these criteria.

Fly pupae were shipped overnight from USDA-ARS Gainesville, Florida and placed immediately in an emergence chamber (Fig 7). At approximately 12:00 pm CDT on release days, the emergence chamber was placed in the appropriate chamber and approximately 10 flies were aspirated into 24-7 translucent vials (Fig 8 and Fig 9). All flies in the emerging chamber were collected into the transport vials in this manner. After all flies were collected. At approximately 1:00 pm CDT the flies were released into attack chambers or actual mounds (Fig 10 and 11). Observations were made on 10 mounds, including the number of flies observed were noted over a two hour period. The fly pupae were shipped May 8 and May 14, 2002 and the emerged flies released May 13 - May 16, 2002 in Pike County. In Bradley Co. fly pupae were shipped October 1, 2002, and October 7, 2002, the merged flies released October 3 - October 12, 2002.

## *Theiohania solenopsae* Release Method

Using protocols developed at USDA-ARS Gainesville, Florida the microsporidian *Theiohania solenopsae* was released in Miller Co. Arkansas.

The site was selected for the number of fly and mounds, and the absence of the microsporidian. Samples were taken from the site in April 2002 and evaluated for the presence of the organism. Pools of 5-20 workers are prepared and observed with a phase contrast microscope for spores and *T. solenopsae* spores were isolated.

Brood was shipped overnight from USDA-ARS Gainesville, Florida September 9, 2002. On Sept. 11, 2002 mounds were evaluated and GPS coordinates of the mounds were taken (Fig 12, Fig 13, Fig 14). On Sept. 12<sup>th</sup>, 5 - 2.5 grams of infected brood were placed into each of 25 mounds within a circular 1/16 acre plot (Fig 15).

**References:** Clemons, J. B., Clemons, J. B., Parker, J. L., Petty, D. W. 1999. Survival patterns of entomopathogenic activity by *Pseudacteon tricuspis* (Diptera: Phoridae) against *Solenopsis invicta* (Hymenoptera: Formicidae). *Journal of Economic Entomology* 92(2): 422-427.

## Results and Discussion

In 2002, releases of both *P. tricuspis* and *T. solenopsae* were made, however, neither of the organisms is currently detectable. In 2002, releases of the organisms were attempted again. Initial data collection suggests that one of the organisms, *P. tricuspis*, may be established in one location in Arkansas. Monitoring for establishment and impact of both organisms will resume this spring.

A slight difference in the percent emergence of *P. tricuspis* pupal shipments for the two releases were noted (Fig 16). Fifty-five percent of the flies shipped for the May release emerged compared to 40% emergence from the October shipment. Knowledge of the potential viability of the flies may impact future release periods, so as to maximize the potential for

The Pike County release site shows that for the spring and summer of 2002 the flies did reproduce (Fig 17) and were present outside of the release area. About 75% of the mounds 50 yards outside and in each direction (North, South, East and West) from the initial release site were positive for *P. tricuspis* during the final evaluation of 2002. However, it is uncertain if the flies survived the winter. It is also uncertain if phorids from the flies released in Bradley Co. survived the winter. Regarding the observation on September 11, 2002 (Fig 17), it should be noted the observations were made approximately 10 hours after sunrise. Pasquero et al. (1996) observed that *P. tricuspis* exhibits a pattern of activity that peaks 7 to 9 hours after sunrise.

*T. solenopsae* was previously released in 1998 near Hope, Arkansas. Although *T. solenopsae* presence was noted following the 1998 release, recent surveys have not detected the pathogen. The 2002 releases were, while similar in fire ant mound density and queen to worker ratio to the 1998 release site, contain a slightly different soil type.

Valuable experience was gained through the interagency cooperation on this project. Participants learned a great deal regarding techniques and potential factors to look for in the success of any future releases. In addition these releases provided valuable outreach opportunities for county agents associated with this project.

Fig. 5. Pike Co. 2002 phorid release site



Fig. 6. Bradley Co. phorid release site

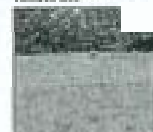


Fig. 7. Emergence chamber



Fig. 8. Transport vial



Fig. 9. Emerging chamber



Fig. 10. Emerging phorids released into attack chambers



Fig. 11. Emerging phorids released into attack chambers



Fig. 12. Emerging phorids directly into a mound



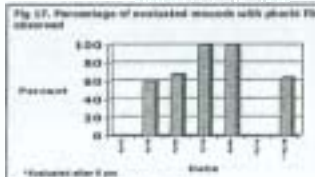
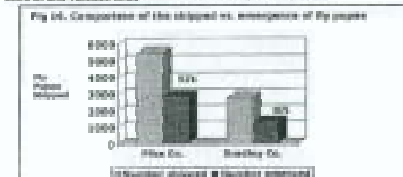
Fig. 12. Aerial view of *Theiohania* release site. Fig. 13. Map of mounds in *Theiohania* release site.



Fig. 14. Map of mounds in *Theiohania* release site.



Fig. 15. 50 and 100 ft buffer surrounding a mound with *T. solenopsae*.



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Thanks to Bill Blevins, USDA-ARS Plant Staff for IAD assistance, the USDA-ARS Plant Staff for the materials used in releases, David St. John, USDA-ARS Gainesville, FL, for the support, Sanford Parker, USDA-ARS Gainesville, FL, for assisting with the release and monitoring, and David St. John for supplying materials and growing flies for the Bradley Co. release.





# Morphological Embryonic Development of the Red Imported Fire Ant *Solenopsis invicta* Buren (Hymenoptera: Formicidae)

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## Abstract

Embryonic characteristics during embryonic development was studied by external observation of the red imported fire ant *Solenopsis invicta*. Early embryonic patterning belonged to the long-germ-type. The germ band formed at the ventral side of the egg, composed of the incipient head lobes, the gnathal and thoracic region, and a growth zone. The germ band developed by extending and curving dorsally in both anterior and posterior directions, until the head and the tail almost connected with each other on the dorsal side of the egg when the germ band reached its maximum length. In the mean time, the segmentation and appendages appeared on the germ band. Five distinctive embryonic stages were evident according to the external characteristics of the growing embryos.

## Introduction

Hymenoptera is one of the best investigated insect orders in the field of embryology. Most of the subjects studied are in the families: Tenthredinidae, Trichogrammatidae, Ichneumonidae, Braconidae, and Apidae. However, little information is available on the embryology of the Formicidae.

The red imported fire ant, *Solenopsis invicta* has become a major pest since its introduction into the United States. Its damage and substantial impact on people, animal, and agriculture are growing because of its aggressive nature. Though intensive studies have been done on its biology, no research has been conducted on its embryology which may help to expand our knowledge of its community structure and management during the early stage of its life cycle.

The present paper reports the external characteristics of *S. invicta* embryos during the process of embryonic development.

## Materials and Methods

Emergent *S. invicta* alates were collected on the campus of Auburn University in May 2002. Alates were kept in petri dishes containing moist paper towel and food source, and maintained in an incubator at 25 °C, 95% RH, in total darkness. Eggs were collected daily two days after alates collection.

The eggs were fixed in Carnoy's solution for about 3–5 minutes before being transferred to 90% alcohol. Egg development was observed and photographed under a stereo-microscope.

Fifteen newly laid eggs were measured for length and diameter at the widest point, using a micrometer and a stereo microscope.

## Results

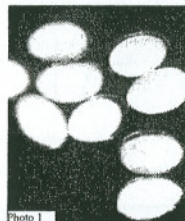


Photo 1. Newly laid eggs are ellipsoid, with a slightly rounded anterior end and pointed posterior end,  $0.45 \pm 0.02$  mm in length and  $0.28 \pm 0.02$  mm in maximal width.

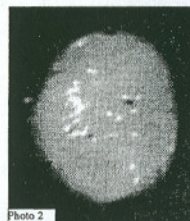


Photo 2. Developmental stage 1: Long germ band forms and develops at the posteroventral side of the egg.



Photo 3 and 4. Developmental stage 2: Germ band extends in both anterior and posterior directions: the posterior end curves around the posterior pole of the yolk mass onto the dorsal surface, and the anterior end immerses anterodorsally into the yolk mass. By this time, the segmentation is completed and the appendages appear.

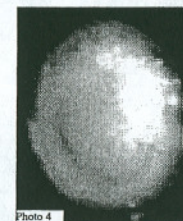


Photo 5 and 6. Developmental stage 3: Germ band begins to shorten and widen with the head and the tail on the dorsal surface moving towards the anterior and posterior pole of the egg respectively. The appendages on the head grows longer, but those on the thorax and abdomen stay unchanged.

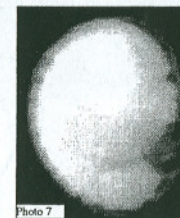


Photo 7. Developmental stage 4: The head of the embryo rotates from the dorsal side to ventral side of the egg and the dorsal closure of the embryo continues.



Photo 8 and 9. Developmental stage 5: The embryo begins to mature after dorsal closure and takes the form of a larva with the head directed backwards. At this time, the larva is ready to hatch from the anterior end of the egg.



## Discussion

- The early embryonic patterning of *S. invicta* was of the long-germ-type, and the embryonic primordium was long and composed of the incipient regions that would produce the head lobes, gnathal parts, thorax, and the abdomen.
- The blastokenesis of *S. invicta* was more distinct than that of other Hymenopteran insects previously investigated. It consisted of three successive embryonic movements: elongation of the germ band, shortening and widening of the germ band, and the displacement of the head anteriorly.
- In Hymenoptera, the extension of the germ band was generally exhibited at the posterior end, but in *S. invicta*, it was proceeded in both anterior and posterior directions. The posterior end of the germ band curved around the posterior pole of the yolk mass onto the dorsal surface, and the anterior end immersed anterodorsally into the yolk mass. The latter is not found in the embryogenesis of other Hymenopterans.
- In other insects, during the process of dorsal closure, it is usual for the embryo to take in all the yolk materials. However, it was interesting that there were some yolk balls left in the outside of the head region, when most of the yolk materials were enclosed within the embryo.



# Release and Establishment of the Fire Ant Decapitating Fly, *Pseudacteon tricusps*, in the Southeastern United States

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Between 1996 and 2002 the decapitating fly, *Pseudacteon tricusps*, was released at 56 sites in the southeastern United States (Fig. 1) as a self-sustaining biocontrol agent of the red imported fire ant, *Solenopsis invicta*. Most releases have been made by the USDA-ARS, CMAVE, Gainesville, FL (32 sites) and the University of Texas, Austin (15 sites). In the spring of 2002, USDA-APHIS funded a joint rearing effort (USDA-ARS, USDA-ARS, and the Florida Department of Agriculture) that has taken over rearing and release responsibilities from USDA-ARS. Overwintering populations of flies were successfully established at 19 sites in 6 states (AL-3, FL-5, LA-3, MS-1, SC-2, TX-5). Fly populations at 10 of these sites have expanded a mile or more away from their release sites (Table). Evaluation of recent releases are still in progress at an additional 19 sites and releases appear to have failed at 16 sites. Fly populations from releases around Gainesville, FL fused in 2001 and were expanding outward at the rate of 10-20 miles a year in the fall of 2001 (Fig. 2). We currently estimate that they are coast to coast in North Florida and beginning to move into Georgia. Fly populations near Auburn, AL and Bonita Springs, FL also have begun to expand explosively. Studies of fly impacts are currently underway in FL, LA, AL, MS, TX, OK, and SC. We are rearing two additional species of decapitating flies (*Pseudacteon curvatus* and *Pseudacteon litoralis*) that should be ready for trial field releases on red imported fire ants in the spring of 2003.

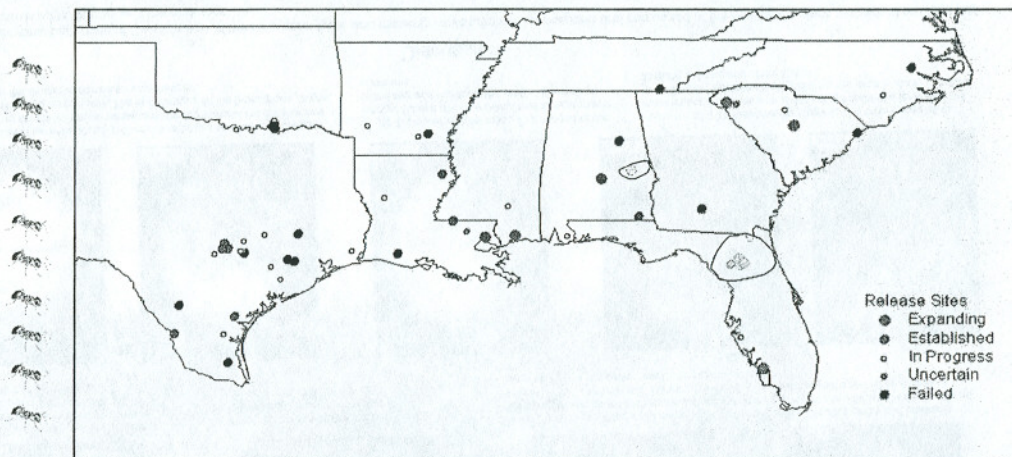


Fig. 1. Releases of the Brazilian decapitating fly, *Pseudacteon tricusps* in the southeastern United States (1996-2002).

State	Release	Source	Pages Used	Status	Expansion
Alabama					
1. Tallapoosa	Aug-98	USDA-ARS	1740	Failed	
2. W. of Auburn	May-99	USDA-ARS	2600	Expanding	12-30 miles
3. Taylor	Sep-00	USDA-ARS	1700	Established	
4. Lowndes	Apr-01	USDA-ARS	—	Expanding	~2 miles
5. Blount	May-02	USDA-APHIS	—	In Progress	
Arkansas					
1. Monticello	Sep-98	USDA-ARS	2400	Failed	
2. Blytheville	May-02	USDA-ARS	5500	In Progress	
3. Warren	Oct-02	USDA-APHIS	2000	In Progress	
Florida					
1. Ft. Gainesville	Sep-97	USDA-ARS	1600	Expanding	20-50 miles
2. near La Crosse	Sep-98	USDA-ARS	1000	Expanding	20-50 miles
3. Gainesville airport	Oct-98	USDA-ARS	1810	Expanding	30-50 miles
4. W. of Newberry	Nov-98	USDA-ARS	1100	Established	
5. Bonita Springs	Nov-99	USDA-ARS	2000	Expanding	5-25 miles
6. Sarasota	Nov-02	USDA-APHIS	—	In Progress	
Georgia					
1. Tifton	Oct-00	USDA-ARS	—	Failed	
Louisiana					
1. Cocodoches	Sep-99	USDA-ARS	2900	Expanding	1-3 miles
2. Norwood	Apr-00	USDA-ARS	1100	Established	
3. Montpelier	Sep-00	USDA-ARS	3500	Uncertain	
4. La. Jolie	Apr-01	USDA-ARS	—	Failed	
5. east of Delhi	Sep-01	USDA-ARS	—	Established	
6. Natchitoches	May-02	USDA-APHIS	—	In Progress	
Mississippi					
1. Senatobia	Apr-00	USDA-ARS	3500	Expanding	5-8 miles
2. Hattiesburg, airport	Apr-02	USDA-APHIS	2500	In Progress	
North Carolina					
1. Oklawaha	May-00	USDA-ARS	5610	Failed	
2. Warsaw	Apr-02	USDA-APHIS	—	In Progress	
Oklahoma					
1. Cato	May-99	USDA-ARS	1900	Failed	
2. Dewar	May-00	USDA-ARS	—	Failed	
3. Dumas	Sep-00	USDA-ARS	2100	Failed	
4. east of Tulsa	Dec-02	USDA-ARS	3000	In Progress	
5. near Cullum	Jan-02	USDA-ARS	3000	In Progress	
6. Rehoboth	Jan-02	USDA-APHIS	—	In Progress	
South Carolina					
1. Cheraw	May-99	USDA-ARS	4810	Expanding	1-2.5 miles
2. Myrtle Beach	Jan-00	USDA-ARS	2000	Failed	
3. Ft. Jackson	Feb-00	USDA-ARS	2300	Expanding	0.5-1.5 miles
4. Pelzer	May-00	USDA-ARS	1000	Uncertain	
5. N. of Columbia	Sep-02	USDA-APHIS	—	In Progress	
Tennessee					
1. R. of Cleveland	Aug-99	USDA-ARS	4100	Failed	
Texas					
1. Austin	1996-1998	State of Texas	—	Established	
2. Laredo	Feb-98	State of Texas	—	Established	
3. Brown	Aug-99	USDA-ARS	5250	Failed	
4. N. of Houston	Sep-99	USDA-ARS	4000	Failed	
5. Valer	Apr-02	USDA-APHIS	—	In Progress	
6. N. of Houston	Apr-02	USDA-ARS	—	In Progress	
7. Houston	—	State of Texas	—	In Progress	
8. Hays Co.	—	State of Texas	—	Established	
9. Matagorda Co.	—	State of Texas	—	Failed	
10. Travis Co.	—	State of Texas	—	Established	
11. Waller Co.	—	State of Texas	—	Failed	
12. Lavi Co.	—	State of Texas	—	In Progress	
13. Travis Co.	—	State of Texas	—	Established	
14. Kibler Co.	—	State of Texas	—	In Progress	
15. Lubbock Co.	—	State of Texas	—	Failed	
16. San Antonio Co.	—	State of Texas	—	Established	
17. Colorado Co.	—	State of Texas	—	In Progress	
18. Wilbarger Co.	—	State of Texas	—	Failed	
19. Warren Co.	—	State of Texas	—	In Progress	

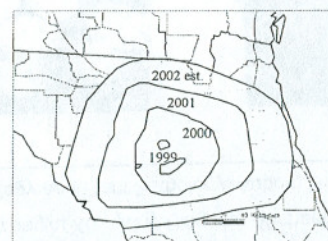


Fig. 2 Expansion of *P. tricusps* from releases around Gainesville, FL (1997-1998).



## Acknowledgements

The releases reported in this poster could not have been done without the help of numerous people in each of the states. Everyone that deserves to be acknowledged cannot be mentioned, but the following people were key to releases in their respective states: Kathy Flanders, Henry Dorough (AL), Lynne Thompson, Donna Shanklin, Kelly Loftin (AR), Lloyd Davis, Cynthia Vann (FL), Wayne Gardner, Stan Diffie (GA), Don Henne (LA), Kathleen Kidd (NC), Wayne Smith, J.T. Vogt (OK), Clyde Gorsuch, Tim Davis, Janet Kintz (SC), Roberto Pereira, Karen Vail (TN), Charles Barr, Jerry Cook (TX).





# Potential Global Range Expansion of the Red Imported Fire Ant, *Solenopsis invicta*

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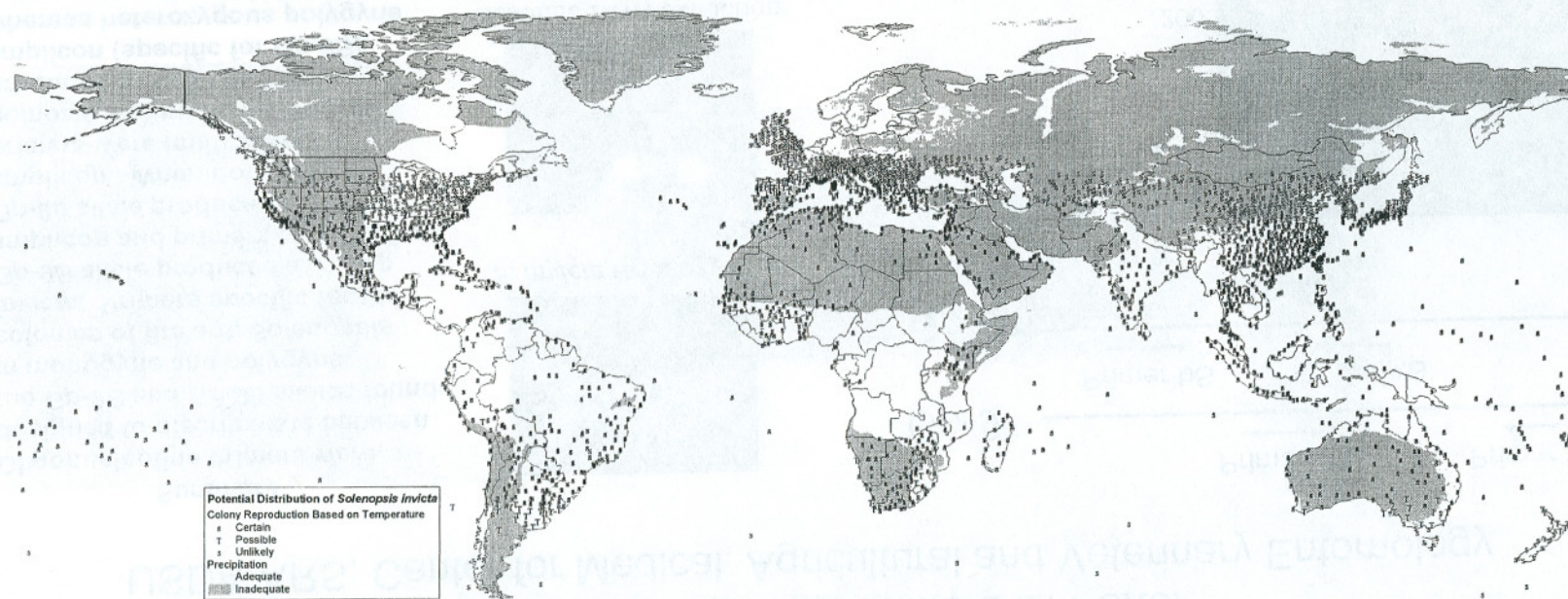
<sup>1</sup>Center for Medical, Agricultural and Veterinary Entomology, USDA-ARS, P.O. Box 14565, Gainesville, FL 32604 USA

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**Abstract:** The red imported fire ant, *Solenopsis invicta* Buren, is an invasive pest that has become widespread in the southern United States and Caribbean after introduction from South America in the 1930's. This species has diverse detrimental impacts on recipient communities. It was recently discovered in Australia and New Zealand and has the potential to colonize numerous other regions. We used a dynamic, ecophysiological model of colony growth (Korzukhin et al. 2001) to predict the potential global range expansion of this invasive species. Based on minimum and maximum daily temperatures, the model estimates colony alate production and predicts future geographic range limits. Because *S. invicta* populations are limited by arid conditions as well as cold temperatures, we superimposed precipitation data upon temperature-based predictions, to identify regions that do not receive enough rainfall to support this species across the landscape. Many areas around the globe, including large portions of Europe, Asia, Africa, Australia, and numerous island nations, are at risk for *S. invicta* infestation. Quarantine officials

should be vigilant for any accidental introductions of this pest in susceptible regions. Costs of eradication increase dramatically as the area of infestation grows, and large infestations may be impossible to eradicate. Other South American *Solenopsis* fire ants (e.g., *S. richteri* Forel) may become invasive if the opportunity arises, and our predictions for *S. invicta* may approximate the potential range limits for these species as well. Manuscript in review in *Biological Invasions*. More detailed maps can be viewed online at [http://cmave.usda.gov/ifa/ifa\\_home.html](http://cmave.usda.gov/ifa/ifa_home.html)

Korzukhin MD, Porter SD, Thompson LC and Wiley S (2001) Modeling Temperature-Dependent Range Limits for the Fire Ant *Solenopsis invicta* (Hymenoptera: Formicidae) in the United States. *Environmental Entomology* 30: 645-655



**Figure Legend.** Symbols correspond to locations of weather stations (n=3,421) providing at least 5 years of reliable data. Red circles indicate areas of certain reproductive success; yellow triangles indicate areas of possible reproductive success; white circles indicate areas of unlikely reproductive success, based on temperature. Green shading indicates regions with sufficient annual precipitation (estimated at >510 mm) to sustain *S. invicta* across the landscape; olive green shading indicates arid regions that likely have insufficient rain (~510 mm annual precipitation). *Solenopsis invicta* will survive, however, in arid regions that are irrigated or near natural water sources.



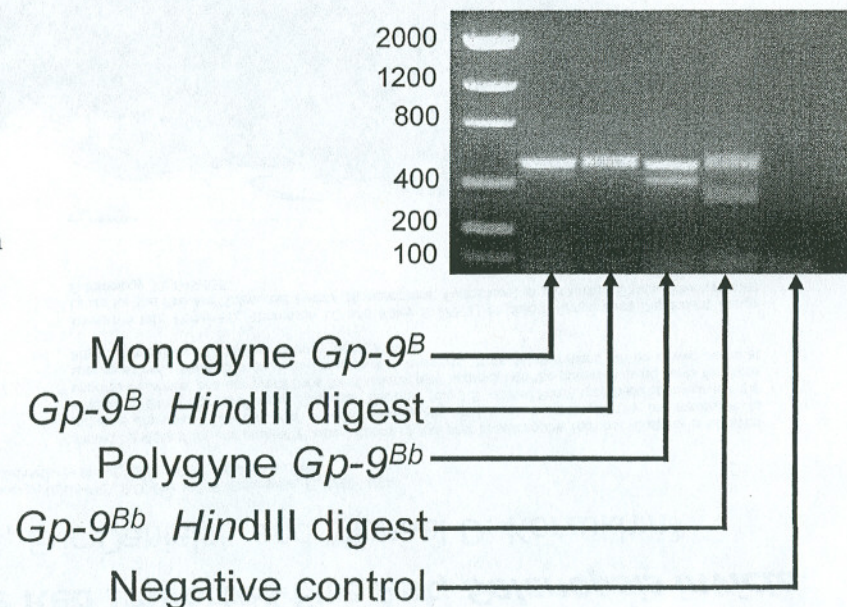
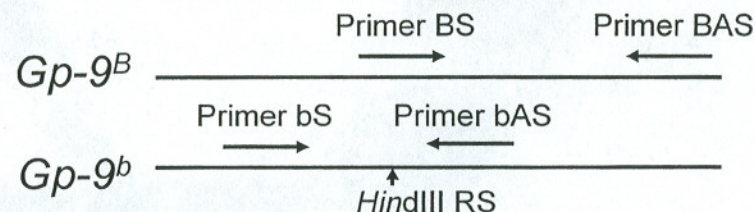
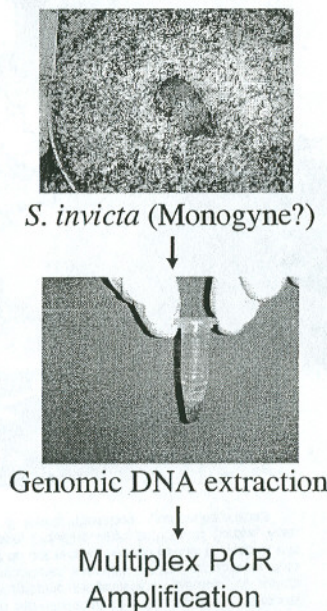
# IDENTIFICATION OF POLYGYNE AND MONOGYNE FIRE ANT COLONIES (*SOLENOPSIS INVICTA*) BY MULTIPLEX PCR OF GP-9 ALLELES

Steven M. Valles and Sanford D. Porter

USDA-ARS, Center for Medical, Agricultural and Veterinary Entomology

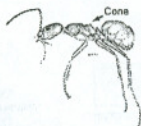
## Summary

Oligonucleotide primers were designed to discriminate between the *Gp-9B* and *Gp-9b* alleles found in monogyne and polygyne colonies of fire ant, *Solenopsis invicta*. Primers specific for the *Gp-9B* allele produced a 517 bp amplicon and primers specific for *Gp-9b* allele produced a 423 bp amplicon. When both sets of primers were multiplexed, homozygous monogyne ants produced a single 517 bp amplicon (specific for *Gp-9B*), whereas heterozygous polygyne ants produced one 517 bp amplicon and one 423 bp amplicon (specific for *Gp-9B* and *Gp-9b*, respectively) which allowed the *Gp-9* alleles to be discerned in a single reaction. This method was tested on ants from 20 monogyne colonies and 20 polygyne colonies and was 100% accurate in discriminating the two forms.





# PRELIMINARY ASSESSMENT OF *DORYMYRMEX* AND *SOLENOPSIS* INTERACTIONS



• Calixto, Marvin K. Harris, Allen Knutson and Charles Barr.  
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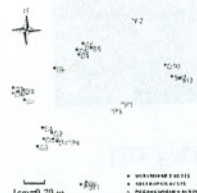


## INTRODUCTION

Hung (1974) made preliminary observations on fire ants (*Solenopsis invicta*) recovered from a single refuse pile of the Pyramid ant *Conomyrma* (= *Dorymyrmex*) *insana* and found fire ants predominated (43%); some authors (Smith 1965; Nickerson et al. 1975; Wilson et al.) suggested *Dorymyrmex* may kill newly mated fire ant queens, alate males and workers, and that perhaps it sometimes nests at the edge of the fire ant mounds, but these observations have not been quantified. The role of native ants in habitats treated for fire ants is not well understood. More knowledge of the interactions of fire ants and native ants is needed. We observed fire ant remains adjacent to *D. flavus* nests in a commercial pecan orchard in Burleson Co., TX while conducting extensive investigations on fire ant in pecan. The *D. flavus* nests were located in plots previously treated with Extinguish® bait (0.5% methoprene applied at 1.5 lbs/acre, on May 19, and October 12, 2000 and June 12 in 2001) and had fire ant nests nearby. Fire ant nest density had been reduced in these plots by about 70%. We documented the observations by collecting the midden (a raised, loosely aggregated mound of earth, sticks, dead ants, ant parts, and other materials constructed at the nest entrance by *Dorymyrmex*) from each of ten nests on July 31, 2002 and again on Aug. 8, 2002. Middens were inspected with the aid of a binocular microscope (4-60x) searching for arthropod remains and other materials that might be of interest. More than 98% of the arthropod remains found consisted of fire ants, with the initial inspection on July 31 showing an average minimum of 203 dead fire ants per midden and the Aug. 8 inspection showing 40 per midden. Further investigations are being conducted to determine how the fire ants came to be in the middens and whether or not these findings are important in fire ant management.

## MATERIALS AND METHODS I

- Study site: Commercial pecan orchard at Mumford, TX (Robertson Co.). (30° 44' 55" N; 96° 33' 12" W).
- 10 middens of *Dorymyrmex flavus* were collected randomly in an area previously treated for fire ants with methoprene bait Extinguish® (IGR) (applied on May 19, and October 12, 2000 and June 12 in 2001).



GPS PLOT OF ANT NESTS

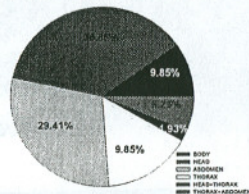
- The entire midden was collected from each nest on July 31, 2002 and nests were marked and referenced with GPS; middens were again collected on August 8 2002 from the same nests.

## MATERIALS AND METHODS II

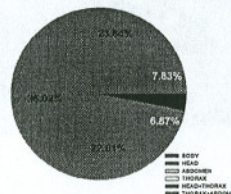
- The middens were placed in plastic snap cap vials (7 dram) and returned to the lab. Each midden was initially divided into 1.5 ml subsamples by volume. Then each subsample was inspected, cleaned of debris and soil particles, and all insect remains were sorted and preserved in alcohol. Arthropods and body parts were placed in vials with 70% alcohol. Ants were identified and counted, with body parts classified as:
  - Body= if body is complete; head-thorax-abdomen
  - Head= if only head is visible
  - Thorax= if only thorax is visible
  - Abdomen= if only abdomen is visible
  - Head + Thorax= if only head and thorax is visible
  - Thorax + Abdomen= if only thorax and abdomen is visible
- Parts of previously identified fire ants and native ants were used to compare and identify remains.
- Time spent on each sample on cleaning, sorting, etc. = 40 minutes
- Time spend on counting remains in each subsample: 45 minutes

## RESULTS AND DISCUSSION I

SOLENOPSIS INVICTA IN TEN DORYMYRMEX MIDDENS  
TOTAL BODIES = PARTS= 4870 (JULY 31 2002)

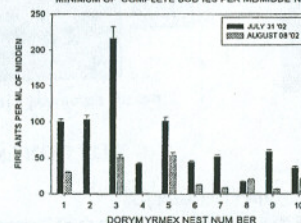


SOLENOPSIS INVICTA IN TEN DORYMYRMEX MIDDENS  
TOTAL BODIES = PARTS= 2145 (AUGUST 8 2002)



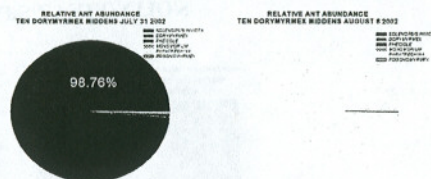
Fire ant segments (separate head, thorax or abdomen) predominated in *Dorymyrmex* middens on both sample days. Whether *Dorymyrmex* ants preyed upon the fire ants or just collected them remains unclear.

SOLENOPSIS INVICTA IN TEN DORYMYRMEX NESTS  
MINIMUM OF COMPLETE BODIES PER MUMMIDE N



Fire ants collected and recovered from each nest (July 31 '02: AVG= 77.24, SE:18.15; August 8 '02: AVG=20.54; SE:6.11)

## RESULTS AND DISCUSSION II



Body remains of five ant species were found on middens; *Solenopsis invicta*, *Dorymyrmex flavus*, *Pheidole* sp., *Monomorium minimum*, *Paratrechina* sp. and *Pogonomyrmex barbatus*. Note fire ants predominated.

## CONCLUSIONS

- *Solenopsis invicta* is the most common species occurring in *Dorymyrmex* middens. Body parts (head, thorax or abdomen) were more abundant than entire ants, but handling may have fragmented intact ants before inspection.
- The differences between sample days suggest that *Dorymyrmex* ants accumulate fire ants at a high rate over a short time period. If they are preying upon fire ants, this may indicate their potential in biological control.
- Other native ant species including *Dorymyrmex* do not occur in significant numbers in the middens.
- Further studies are necessary to understand relationships among ant species and evaluating their potential as biological control agents. When combined with bait applications, *Dorymyrmex* seems to tolerate bait treatments (being little affected by the IGR) and second by competing with fire ants.

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## Acknowledgments

- Dash Bhagirathi provided lab help.
- Partial Funding was provided by the TEXAS FIRE ANT INITIATIVE and USDA Special Grant

More information about FIRE ANTS

• <http://fireant.tamu.edu>





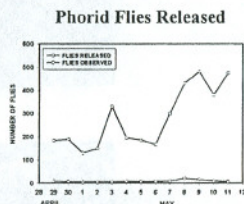
# AREAWIDE SUPPRESSION OF FIRE ANTS - TEXAS HIGHLIGHTS OF YEAR 2002



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c-barr@tamu.edu

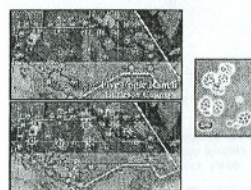


## Five Eagle Ranch - Burleson Co. BIOLOGICAL CONTROL SITE



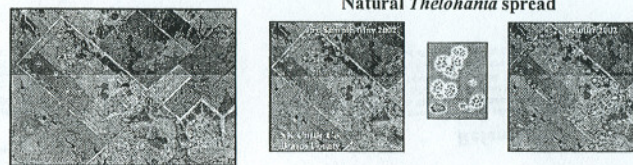
3,895 flies were released between April 29 and May 11. 100 flies observed following days of releasing. 3 recoveries on July 29.

### Natural *Thelohania* spread



## NK Ranch Co. - Brazos Co. CONTROL SITE

### Natural *Thelohania* spread



## MOUND COUNTS (Treatment applied on May 31)



1475 mounds; AVG=21.6; STD: 14.73. **Treated** (0 photo): 538 mounds; AVG=26.76; STD: 13.13. **Control** (0 photo): 940 mounds; AVG=31.33; STD: 15.43.

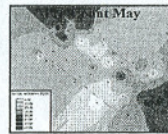


421 mounds; AVG=9.23; STD: 8.30. **Treated** (0 photo): 44 mounds; AVG=1.2; STD: 4.25. **Control** (0 photo): 397 mounds; AVG=13.23; STD: 7.7.

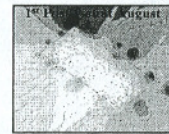


630 mounds; AVG=12.4; STD: 11.47. **Treated** (0 photo): 42 mounds; AVG=1.1; STD: 2.40. **Control** (0 photo): 588 mounds; AVG=19.6; STD: 9.57.

## MOUND COUNTS (Bait applied on October 11)



1264 mounds; AVG=24.08; STD: 18.14. **Treated** (0 photo): 198 mounds; AVG=13.75; STD: 6.94. **Control** (0 photo): 809 mounds; AVG=34.96; STD: 16.08.



484 mounds; AVG=8.68; STD: 7.7. **Treated** (0 photo): 40 mounds; AVG=1.0; STD: 3.09. **Control** (0 photo): 364 mounds; AVG=12.13; STD: 7.31.



807 mounds; AVG=16.14; STD: 16.20. **Treated** (0 photo): 180 mounds; AVG=4.65; STD: 10.45. **Control** (0 photo): 706 mounds; AVG=23.53; STD: 11.09.

## FORAGING EVALUATION



2479 Fire Ants apprx.; AVG=40.50%; STD: 2.48. **Treated** (0 photo): 1203 IFA foraging; AVG=42.63%; STD: 13.49. **Control** (0 photo): 1426 IFA; AVG=47.54%; STD: 24.33.



1764 Fire Ants apprx.; AVG=14.89%; STD: 26.18. **Treated** (0 photo): 105 IFA foraging; AVG=2.54%; STD: 9.79. **Control** (0 photo): 1699 IFA; AVG=43.22%; STD: 12.44.

## FORAGING EVALUATION



2188 Fire Ants apprx.; AVG=55.74%; STD: 13.24. **Treated** (0 photo): 1118 IFA foraging; AVG=55.30%; STD: 13.63. **Control** (0 photo): 1470 IFA; AVG=56.68%; STD: 12.95.



3099 Fire Ants apprx.; AVG=41.80%; STD: 31.20. **Treated** (0 photo): 232 IFA foraging; AVG=11.69%; STD: 19.86. **Control** (0 photo): 1858 IFA; AVG=61.30%; STD: 19.45.

## PITFALL TRAPS EVALUATION

Total of 26,825 IFA captured on May; AVG=67.06; STD:108.44. 9 Species recorded being IFA the dominant (99.47%), followed by *Diplophoropium* (0.281%), *Paratrechina* (0.177%), *Dorymyrmex* (0.10%), *Tapinoma*, *Formica*, *Pachycondyla*, *Pogonomyrmex* and *Monomorium* sharing (0.003%).



## PITFALL TRAPS EVALUATION

Total of 12,618 IFA captured on May; AVG=31.54; STD:49.74. 5 Species recorded being IFA the dominant (99.76%), followed by *Monomorium* (0.012%), *Paratrechina* (0.09%), *Pogonomyrmex* (0.007%) and *Hypoponera* (0.007%).





# The use of community participation in surveillance for RIFA in South East Queensland, Australia



Michelle Milzewski, Manager Community Engagement  
Jenny Bibb, Manager Public Relations  
Fire Ant Control Centre Brisbane – Australia

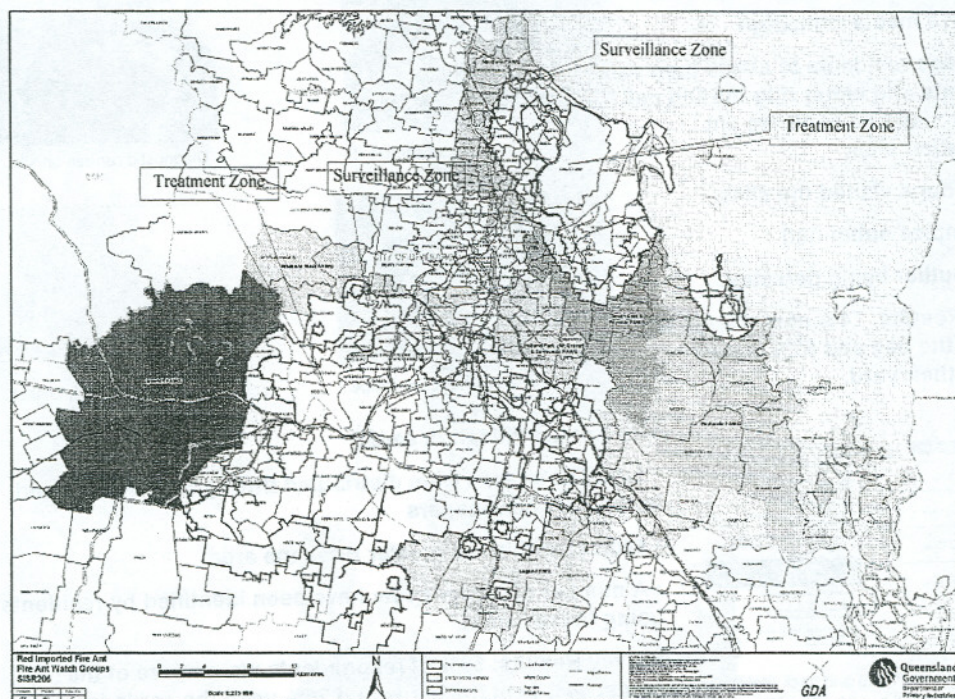


**Challenge:** To find any undetected fire ant nests outside the existing Treatment and Surveillance Zones.

**Fire Ant Community Watch Group Initiative** – a partnership between government and community.

Seven groups are established across the greater Brisbane area with more to follow. Groups are comprised of individuals and representatives of other community and environmental groups. FACC provides training, secretarial and coordination support. Groups conduct surveillance in parkland areas and assist in maintaining community support through public education.

Map showing areas covered by each Fire Ant community watch group in relation to the treatment and surveillance zones of the fire ant eradication program.



Over 50 parkland areas have been checked for fire ants.



Fire ant watch groups maintain community awareness and support.



There are over 180 Fire Ant Rangers.



# The use of public relations activities in passive surveillance for RIFA in South East Queensland, Australia



Jenny Bibo, Manager Public Relations  
Michelle Milzewski, Manager Community Engagement  
Fire Ant Control Centre Brisbane – Australia



**Challenge:** To find any undetected fire ant nests outside the existing Treatment and Surveillance Zones.

Passive surveillance promoted and encouraged through public relations activities including distribution of Identification cards and Find the Fire Ant Day.



**Find the Fire Ant day – 28 July 2002**

- extensive media campaign
- encourage residents to check their yard for fire ants and either ring the DPI call centre or visit one of 10 fire ant identification sites
- 2609 groups visited the sites,
- 341 samples submitted
- 1 new outlier being detected.

**Survey Results:** 74% of residents were aware of the day and 52% actually checked their yard.



Information and sample ID site.



Diagnostic centres on site.



Extensive media coverage.



Map showing 10 info sites.



Fire Ant ID card front (left) and back (right). Actual size 10.5 x 7.5 mm (4 x 3 inches).

## Fire Ant identification cards

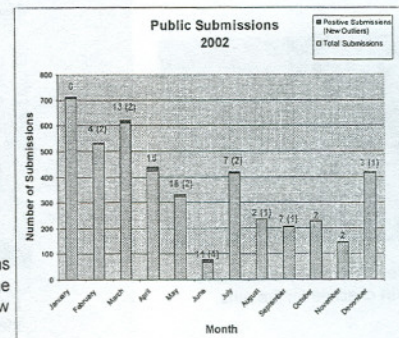
- 1.4 million cards were distributed throughout Queensland in regional newspapers
- 540,995 within the greater Brisbane area.
- A number of outlier nests have been identified by residents using these cards.

**Survey Results:** 59% of respondents were aware of the cards, 44% kept the cards and 26% used the cards to check their yards.

## Passive Surveillance program results:

- 27 outliers detected outside the treatment zone
- 15 outliers detected by public
- 7638 sample submissions by public
- 72,500 calls to call centre
- 85,500 visits to website

Fire Ant sample submissions by the public in 2002 and the number of positives and new outliers found.





# Diagnostics & its role in the fire ant eradication program in Brisbane - Australia.



Marlene Elson-Harris, Kym Johnson, Shane Moloney & Lynne Griffin  
Fire Ant Control Centre Brisbane – Australia



Diagnostics underpins all operational units within the Fire Ant Control Centre with accurate and timely identifications.



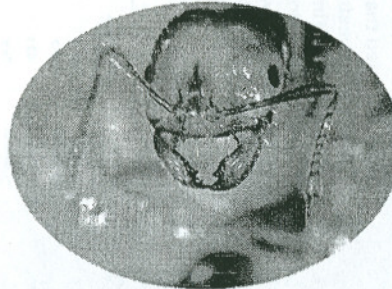
On-site identifications at various centres throughout the greater Brisbane Area.

Entomological training for community watch groups.



Spin-offs from Diagnostics:

Early detections & eradication of otherwise undetected incursions including Argentine ants *Linepithema humile*, Tropical fire ants *Solenopsis geminata* (above left) and Crazy ants *Anoplolepis gracilipes* (above right).



All ant samples generated by the program are identified by the diagnostics unit.

Since February 2001 > 31400 samples have been identified.

To date 9% of these have contained *Solenopsis invicta*.

Up to 1200 samples per week have been processed by this unit.

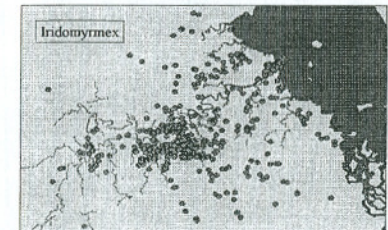


*Solenopsis invicta*



Providing scientific support for public relation events.

Ensuring accurate scientific information for community engagement meetings.



This comprehensive survey has enhanced our knowledge of native ant distributions in south east Queensland. Currently 52 of 103 known Australian genera have been recorded.



# Increasing the treatment options for fire ants in Brisbane - Australia



John R. Hargreaves & John Johnston  
Fire Ant Control Centre Brisbane – Australia



Specific niches have occurred within the treatment programme that have allowed us to expand our treatment options.

## Hay bales

Although methyl bromide is registered for use against ants at 120g/bale for mulching straw or hay, there was no registration for hay in storage for fodder. However, the general fumigation rate of 32g/cubic meter gave excellent control of RIFA. The National Registration Authority (NRA) has ratified this use as permit No 5168.

Sequence	% Mortality	% Efficacy at 95% Confidence Level
Bale insertion without fumigation	2.3	
Fumigation 9 – 10 Oct 01	100	99.84
Fumigation 11-12 Oct 01	100	99.69
Fumigation 16 – 17 Oct 01	100	99.78
Total Fumigated	100	99.93

Sampling interval Pasture	Site 1	Site 2	Site 3	Site 4	Mean and Standard error
2 hrs after	0.364	0.103	0.302	0.047	0.204±0.066
1 day	1.332	0.910	0.000*	0.172	0.603±0.271
3 days	0.581	0.379	0.922	0.079	0.490±0.153
7 days	0.318	1.365	0.000*	0.104	0.420±0.280
14 days	0.028	0.095	0.042	0.000*	0.041±0.017
28 days	0.000* **	0.000* **	0.000*	0.000*	0.000*
Soil					
2 hrs after	3.148	0.484	0.079	0.087	0.949±0.640
1 day	0.670	0.746	0.093	0.504	0.503±0.126
3 days	0.712	0.344	0.000*	0.051	0.277±0.095
7 days	1.196	0.123	0.008	0.000*	0.332±0.251
14 days	0.103	0.019	0.000*	0.009	0.033±0.021
28 days	0.010 **	0.000* **	0.000*	0.000*	0.000*

S-methoprene residues expressed in mg/kg dry weight from grazing locations within the treatment area for fire ant baiting.

## S-methoprene residues on pastures

The study provided valid S-methoprene residue data to the National Registration Authority (NRA) for the treatment of pasture. A suitable withholding period for grazing beef cattle could consequently be established. It was calculated that 1mg S-methoprene / kg dry weight of pasture as 100% of a beast's diet was the maximum feeding rate. The data from the trials were submitted to NRA. A permit, (No.5998) was issued allowing a nil withholding period for grazing animals.

## Protective treatment for nursery stock

A slow release formulation of chlorpyrifos is used in the nursery trade as a control for beetle larvae (black vine weevil) at the usage rate of 1kg/m<sup>3</sup> potting mix. We know from previous trials that RIFA workers can be controlled at 10-12mg chlorpyrifos / kg potting mix. The table (right) shows the decay in a production glasshouse over 6 months. An application for an interim permit for 6 months protection has been forwarded to NRA. Data to date still shows levels of 42 – 51mg / kg in mix exposed for 12 months.

Date Tested	Chlorpyrifos mg/kg	% Mortality suSCon	% Mortality Untreated
30Jan02	61.0	100	6.7±0.4
20Feb02	87.2	100	4.7±2.2
20Mar02	102.1	100	4.2±2.2
19Apr02	78.9	100	4.1±1.7
22May02	82.6	100	1.1±1.5
21Jun02	83.6	100	2.1±1.1
17Jul02	51.3	100	7.0±0.8
22Aug02	109.8	100	5.9±1.03

Residues and mortality of RIFA workers exposed to the collected mixture.

Treatment	Conc	Pretreat	3DAT	7DAT	14DAT	21DAT
nil		2.8a	2.5a	2.7a	2.7a	2.8a
fipronil	1L / nest	2.8a	2.8a	2.6a	2.5a	2.6a
fipronil	2L / nest	2.8a	2.6a	2.0a	1.4b	1.4b
fipronil	4L / nest	2.7a	1.5b	0.8b	0.4c	0.1c

Treatment	Conc	Pretreat	1DAT	2DAT	3DAT	7DAT	14DAT
nil		3.0a	3.0a	3.0a	3.0a	2.9a	2.9a
chlorpyrifos	20ml / 100L	2.9a	0.1b	0.0b	0.0b	0.0b	0.0b
fipronil	2.5ml/100L	2.9a	2.4c	1.4c	1.0c	0.7c	0.1b

## Fipronil

Trials of fipronil as a drench have shown it to give similar levels of control to the chlorpyrifos standards. Reducing the volume of wash of fipronil from 4L / nest has been significantly less effective. Application for use as a nest injection and drench has been made to NRA.

Mean activity rating per nest after 4L wash had been poured on the nest and in a 1m diameter circle around the nest. Work conducted at Swanbank Qld, October – November, 2002.

## Compost treatment of infested soil

Recordings of temperatures in composting heaps of commercial facilities have shown that some reach a consistently high temperature (over 65 degrees C), very rapidly. With good storage facilities, this treatment is being considered as a RIFA disinfestation procedure.





# Progress of the Fire Ant Eradication Program in Brisbane - Australia



Craig Jennings, Technical Services  
Fire Ant Control Centre Brisbane – Australia

## Treatment

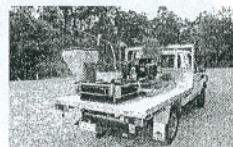
Our aim is to bait the treatment area (green zone on map) 12 times over three years

We are up to the 7<sup>th</sup> round

The treatment area is 41 957 hectares  
102678 land parcels

This has involved 89 586 consents from  
residents to enter their properties

Only 605 residents have refused treatment –  
these undergo continual surveillance



Aerial application: 53%  
All Terrain Vehicles application: 22%  
Manual/foot application: 25%

180 259 hectares have been treated since the beginning of  
the program - this includes 363 963 land parcels

75% of properties had no active nests during a survey of  
900 known infested premises

Monitoring sites show a 90% overall reduction in nest  
density

Large areas of the treatment zone have been free of RIFA  
since February 2002



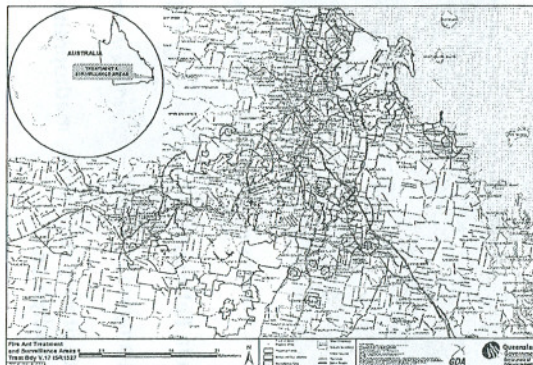
## ERADICATION

The aim of the Fire Ant Control Centre is to eradicate  
Red Imported Fire Ants

They were first discovered in Brisbane Feb 2001

Program commenced 24 September 2001

The eradication program involves a multidisciplinary  
approach that includes treatment, surveillance, PR,  
community engagement, risk management and security  
and science



### FACC Eradication Plan Schedule

	Year					
	2001	2002	2003	2004	2005	2006
Surveillance						
Passive						
Active						
Post treatment						
Treatment						
Scheduled						
Nuisance						

Community based surveillance

Outside the Treatment Zone

Within the Treatment Zone

3-4 Treatments per year

For persistent colonies

## Surveillance

Our aim is survey all land in the surveillance zone (Yellow zone on  
map) at least once a year

Enhanced surveillance zones (Orange zone on the map) around  
outlying infestation will be done twice a year

Outside of the surveillance zone we will be undertaking targeted  
surveillance (based on probability of infestation)

Targeted surveillance will include areas such as new housing  
developments



44 289 hectares have been surveyed outside the treatment  
zone

This area consists of 136 308 land parcels including 112 165  
properties

73 infested properties in 27 outliers have been detected beyond  
the treatment zone

In the 100% surveillance zone the occurrence of infested  
properties is 0.031% or <1 in 3000

60 of these infested properties have fewer than 5 nests

10 outliers are beyond the 100% surveillance zone

We believe that we are close to delineating the extent of the  
RIFA infestation in Queensland





# Ecological effects of *Solenopsis invicta* in Brisbane - Australia



Tania Fuessal†, Adriana Najar†, Jo-anne Holley\* and Kris Plowman\*  
Fire Ant Control Centre Brisbane – Australia



## Assessment of ecological effects of *Solenopsis invicta* and the treatment program on soil and litter invertebrates

Soil invertebrate and mobile ground invertebrate assemblages were sampled in 2002 in sites:

- within the treatment zone with *S. invicta* - TFA
- within the treatment zone and not colonised by *S. invicta* - TNFA
- outside the treatment zone - Control

Treatment began in September/October 2001.



Examples of the sites sampled are shown above.

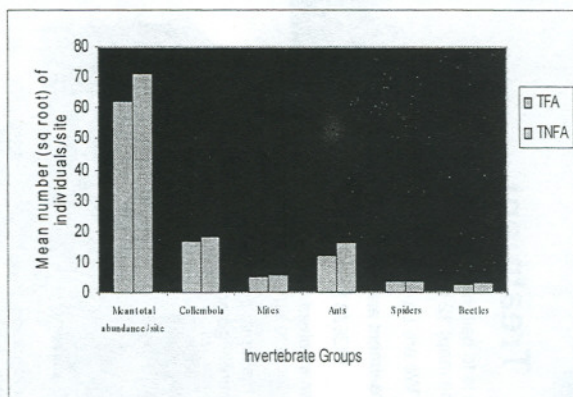
Soil cores (5 x 5 cm) taken in February and extracted by Tullgren funnels provided samples of soil invertebrates.

Mobile ground fauna was sampled by pitfall traps (10 x 7 cm) between May and July.

Treatments included: insect growth regulators, methoprene & pyriproxyfen; and the metabolic inhibitor, hydramethylnon.

Preliminary results suggest that the abundance of mobile ground dwelling invertebrates tends to be greater in treated areas without fire ants than in treated areas with fire ants.

The abundance, diversity and functional groups of beetles and ants recorded in the pitfall traps, and the abundance and diversity of soil dwelling Oribatid mites, is being estimated. These estimates are used to assess the impact of fire ant infestations and the treatment regimes on invertebrate assemblages.



† Tania Fuessal (Griffith University); Adriana Najar (University of Queensland); \* Ecology, Scientific Services, Fire Ant Control Centre.



# Monitoring the fire ant eradication program in Brisbane - Australia

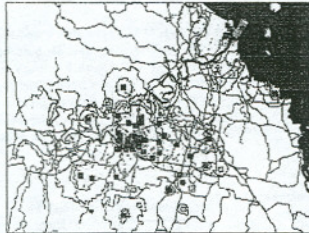


Evan Harris, Stuart Mutzig & Paul Garland  
Fire Ant Control Centre Brisbane – Australia

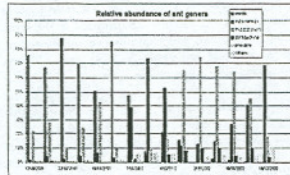
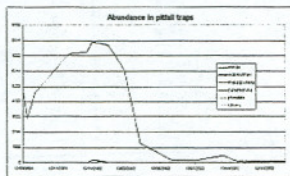


Post treatment monitoring and assessment of treatment regimes throughout infested areas using population monitoring techniques and brood/nest assessments.

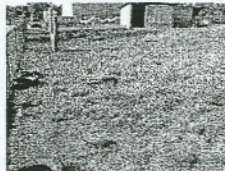
Population monitoring to assess the numbers, nest density and rate of decline in active infestations. Currently 66 sites are regularly assessed.



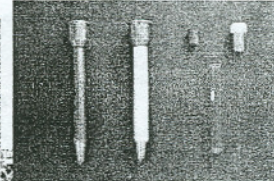
Current distribution of monitoring sites.



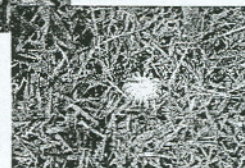
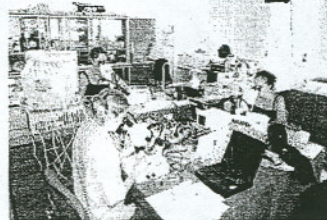
Typical data analysis of a monitoring site.



Semi-rural monitoring site.



Pitfall trap components.

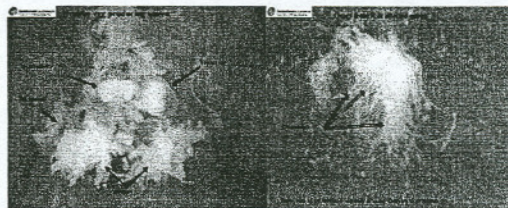


**Brood and nest assessment:** This technique is used to determine the reproductive status of active nests which reveals the effects of the treatment on these areas where population data and surveillance techniques cannot. Presence of worker ants may not necessarily indicate a healthy nest capable of producing viable alates and continuing the infestation.



Active nests are opened and the brood searched for presence of worker pupae. Queens and dealates are harvested for dissection and the presence of other invertebrates in the nest are recorded.

Dissection indicates the insemination and ovary status of those harvested allowing a rating of their viability to be made.



An area of major infestation in suburban Brisbane showing locations of positive RIFA.



The same area showing extent of infestations revealed by property survey.



The area again overlaid with brood assessment results.



## Pastoral Peace?

### A third year report on *Thelohania solenopsae* in a Mississippi coastal pasture.

S. James<sup>1</sup>, A.M. Callcott<sup>1</sup>, H. Collins<sup>1</sup>, D. Oi<sup>2</sup>, R. Weeks<sup>1</sup>, and D. Williams<sup>2</sup>

<sup>1</sup>USDA, APHIS, PPQ, CPHST, Soil Inhabiting Pest Laboratory, Gulfport, MS<sup>2</sup>USDA-ARS/CMAVE, Gainesville, FL.

## INTRODUCTION

In many instances it is not economically feasible or chemically toxic to achieve fire ant suppression. Inferred agricultural trends, such as farmers often fall into this category and thus need to reap great benefit from the introduction of biological controls for imported fire ants. The reintroduction of *Trichobothriopsis* (Monobothrius) *Trichobothrius*, documented inferring and imported fire ants (*Solenopsis invicta*) in Brazil (Rosa et al. 1977) and black imported fire ants (*S. ghoshii*) in Argentina, that have demonstrated by USDA, ARS, ORAFA to decrease colony populations and reduce colony biomass in 1994, 1995, 1996. Therefore, field trials have been initiated to determine the potential of *T. trichobothrius* as a biological control agent here in the United States of America.

## OBJECTIVE

Our objective was to assess *Thelohania* as a biological control agent for *Schizopala* in the southern Mississippi.

## MATERIALS AND METHODS

October 21, 1969 we initiated ARES with the initiation of a trial to evaluate field releases of the polyphagous Thaumaspis subgenus. Two sites in southern Mississippi, one polyphagous in Hancock Co. and one monophagous in Harrison Co. were selected for the bioassay trials. The monophagous site, however, was lost only a day or so before implementation. The four plots at each site were divided into two quadrants with two rows quadrants across plots. Due to the large density of *Thaumaspis* at the polyphagous site, the cluster within plots were kept  $10 \times 10$  m in size instead of the standard  $5 \times 5$  m size. In addition, 2 *T. autumnalis* (field collected by ARES prior to study) was introduced in 100 g amounts in nine quadrants within each of the bioassay plots. An weather and environmental permit, plus evaluations consisting of number, population index, and within-plot location of insects have been conducted every five months. Evaluations to date occurred on weeks 8 (pre-implementation), 12, 20, 28, 36, 44, 52, 78, 84, 91, 100, 108, 116, 127, 140, 155, 161, and 173. Repeated measures ANOVAs, with treatment, year, and season nested within year, year performed in both cluster number and population index. Population indices with insect pairs scaled for a heavier weighting than other variables. Data on sex, feeding from nuptial process, function and fecundity, *Lt* in weeks 152 and 155, and data from weeks 162–164 are analyzed.

Working samples were also collected from each pond during these evaluations and frozen until they could be examined. After being each sample was ground in a flammé grinder and wet faecal slits were made of the resulting slurry. The slides were stained with 40% fastgreen as a phase contrast microscope for presence of *T. polydora* species (Stuart et al. 1984).

## RESULTS and DISCUSSION

## College Magazine

[illegible]

Unmanipulated and watered pine displayed similar current numbers and population indices for the first year of the trial and at some points in the second year. However, though currently *P. rufescens* infections occurred by the third year in some manipulated plots in contrast from the unmanipulated cycling soil. Asynchronised by the current pine (year effects was,  $F_{8,232}=0.044$ ) (manipulation effects was,  $F_{1,232}=17.94$ ,  $P<0.001$ ). Very low infection were found in the area after the flooding that occurred a few weeks prior to the week 156 sampling. As of week 172 the number of current pine colonies had not returned to the level experienced for their first year. We did not monitor asynchronising at that through 2005, but it is reasonable that the floods during end of 2003 have disrupted the current pine at this site beyond further use.



### Declaration of Conflicting Interests

Two aquaria were designed for germinating and rearing (between 21 and 30 weeks) up to larvalization, 2 remains in one of the bioenclosed plots were positive for the spores. By week 20, both of the bioenclosed plots laid down positive vegetation, and spores were discovered from 2 remains in one of the exposed plots. Since that week, no other spore number samples have been fixed for the control plots. The percent of individual colonies in bioenclosed and polymeric plots increased as the number of colonies decreased (Figure 2). Highest numbers of spore positive colonies occurred in the third year while the bioenclosed plots were standing. Since the seasonal temperature increase in the summer.



Among the properties of an ideal biological control agent is the ability to maintain itself in the field. Persistence of infection does appear to be the case, as colonies maintained in the nest chamber will self-renew on new hosts, negative on themselves, and their progeny again have the potential to colonize the original host. Ability to defend themselves against predators and parasites, and to defend themselves against the effects of weather and other environmental factors are also of the utmost importance. In the case of the parasitoid, the host is the egg of the pest, and the parasitoid must be able to find the pest egg, to penetrate the egg shell, to find the pest embryo, to feed on the pest embryo, and to develop within the pest embryo. The parasitoid must also be able to survive in the field, to find the pest egg, to penetrate the egg shell, to find the pest embryo, to feed on the pest embryo, and to develop within the pest embryo. The parasitoid must also be able to survive in the field, to find the pest egg, to penetrate the egg shell, to find the pest embryo, to feed on the pest embryo, and to develop within the pest embryo.



Even in this limited sampling our results do not support the assumption of *Toxotrichia micropus* as a useful biological control. Within three years, significant reductions of dispersal for its population and ability to induce an infection have been demonstrated. The another conclusion and finding of this and some methods for the maintenance of this experiment it was initially designed, but there are typical of the most and fewer monitoring, regardless of the continued status of the research plots, is certainly warranted.

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- Stark, J., D. H. Olsen and J. M. Hall (1998): The translocator complex of the endoplasmic reticulum. *Biochem. Biophys. Res. Commun.* 245: 1-6.



**Table 1, cont.** Red imported fire ant mounds per 0.25 acre circle plot, Lago Santa Fe, Galveston Co., Texas, treated with the hopper blend of Extinguish™ (s-methoprene) and ProBait™ (hydramethylnon) fire ant baits (0.75 lb each product blended together and applied using ground application equipment) on April 18, 2002.

**Number of red imported fire ant mounds/0.25 acre**

<u>Lot Number</u>	<u>July 17, 2002 (12 WAT)</u>	<u>September 26, 2002 (23 WAT)</u>
<b>Treated area:</b>		
32	2	13
25	5	14
24	8	20
23	8	17
20	14	7
2	3	23
1	4	26
46	5	11
Mean $\pm$ Stand. Dev.	6.13* $\pm$ 3.83	16.38* $\pm$ 6.37
<i>T</i> =	10.5679	6.6994
<i>n</i> = 8; d. f. = 8; <i>P</i> =	0.0000	0.0000
Percent reduction:	-85.50%	-61.23%
<b>Untreated area (plot):</b>		
1	-	24
2	-	39
3	-	15
4	-	15
Mean $\pm$ Stand. Dev.		23.25** $\pm$ 11.32
<i>T</i> =		-0.1940
<i>n</i> = 4; d. f. = 6; <i>P</i> =		0.4263
Percent reduction:		

\* Mean significantly different ( $P \leq 0.05$ ) from pre-treatment mean using the Student *T* test (Microstat).

\*\* No significant reduction in mean number of fire ant mounds per plot



## Mitigation of Bifenthrin in Nursery Runoff

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One of the mandatory quarantine measures at nurseries is to treat planting media with Talstar®, a product that contains the active ingredient bifenthrin – a fourth-generation synthetic pyrethroid. However, monitoring by the California Department of Pesticide Regulation (CDPR) has shown that bifenthrin consistently appears in runoff from some nursery sites. Bifenthrin is very stable in the environment and in addition, is extremely toxic to many aquatic organisms including fish and invertebrates. Therefore, preventing the discharge of bifenthrin from nurseries is of great importance for protecting aquatic ecosystems such as urban creeks and streams.

### Experimental

Since 1999, researchers from the University of California and CDPR, in collaboration with scientists from the California Department of Food and Agriculture (CDFA), the Regional Water Quality Control Board, Orange County Public Facilities and Resources Department (OC PFRD), and the nursery industry, have been developing various low-cost practices to reduce pesticide runoff from nurseries. The field site is a typical commercial nursery occupying 120 acres with production of both outdoor and greenhouse container grown plants. Runoff occurs as a result of irrigation and winter precipitation (13 inch yearly average). The low-cost mitigation practices include sediment traps, a sediment pond, and a vegetative filter (Fig. 1). Polyacrylamide (PAM), a long-chain anionic polymer causing flocculation of sediment particles, is the most recently added mitigation practice.

At the site, runoff is directed through sediment traps into a small sediment pond. Runoff then flows through a 260 m channel before exiting the property. The vegetative strip occupies about two thirds of the channel, and consists of canna lilies grown in wire baskets that are suspended in the water. The plants are harvested periodically for sale. The sediment trapped in the sediment traps, sediment pond, and channel is cleaned whenever significant buildup is seen, and then added to container soil used for new plantings.

Pesticides, including bifenthrin, are monitored at the beginning and the end of the plant strip. Whole water samples of 500 mL are extracted by shaking with ethyl acetate in separatory funnels for three times, and the extract is further concentrated and then quantified by analysis using GC-ECD. In addition, runoff and sediment samples were also taken for experiments to determine the distribution of bifenthrin between solids and water in runoff and adsorption coefficient  $K_d$  in sediment.



## Results and Discussion

Bifenthrin enters the runoff and moves with soil/sediment particles. Our mitigation practices operate on the principle of on-site sediment/soil removal and containment. Sediment traps cause physical removal of soil/sediment particles in the runoff. In the sediment pond, the flow rate of runoff is dramatically reduced, which provides time for the suspended solids in the runoff water to settle out. Addition of polyacrylamide (PAM) causes aggregation and subsequent settling out of suspended solids. Water flow is further slowed in the channel by canna lilies that have extensive root systems, which facilitates the separation of suspended solids from the moving water column. It was observed that the upstream runoff at the nursery site consistently contained high levels of solids. The suspended solid content rapidly decreased when the runoff traveled downstream through the sediment trap, sediment pond, and finally the vegetated channel. The greatest drop in suspended solid content occurred between the PAM delivery point and the pond, or after the sediment trap. When the PAM delivery point is used as the reference point, the suspended solid removal after the sediment trap was >90%. More reductions further occurred in the vegetated channel. When the runoff reached the end of the vegetative strip (240 m from the pond), the overall suspended solid removal was 99.6%. The reduction in total mass of suspended solids in runoff was greater than indicated by the sediment concentration data above. The total mass of suspended solids in runoff is equal to the product of runoff volume and suspended solid concentration. Consequently the decrease in runoff over the last few years has further reduced sediment movement off-site relative to years before irrigation and pesticide BMPs were implemented.

Runoff samples were also analyzed for bifenthrin. Bifenthrin concentrations in runoff generally decreased as the runoff moved through the sediment trap, pond, and the discharge channel (*Figure 4*). For instance, on 05/16/2002, the initial bifenthrin level in runoff before the PAM release point was  $10.6 \mu\text{g L}^{-1}$ , which decreased to  $0.87 \mu\text{g L}^{-1}$  at the end of the vegetative strip. On 06/16/2002, the initial bifenthrin level was  $3.2 \mu\text{g L}^{-1}$ , which decreased to  $0.28 \mu\text{g L}^{-1}$  at the vegetative strip. Using the concentration before the PAM release point as the reference point, the reduction in bifenthrin concentration in the runoff was 91.8% on 05/16/2002, and 91.3% on 06/16/2002 (*Table 1*). The greatest decrease occurred after the sediment trap, but further decreases occurred through the vegetated channel. The pesticide removal was apparently correlated with the removal of suspended solids caused by the various BMPs along the runoff path.

In conclusion, our study shows that the use of low-cost mitigation practices can effectively decrease the loading of pesticides from nursery runoff. Our findings on bifenthrin are especially significant, because synthetic pyrethroids are considered as replacements for organophosphate insecticides such as diazinon and chlorpyrifos. Prevention of discharge of these products into surface streams is of great importance in maintaining the health of affected water bodies.

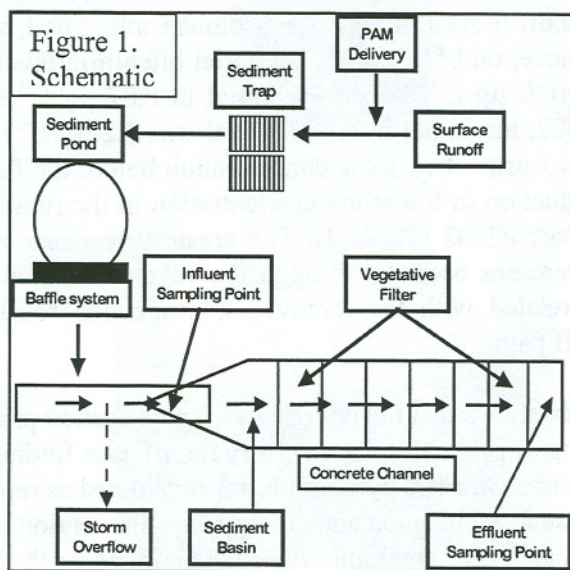


Table 1. Reductions in bifenthrin level in runoff along the runoff path (% of level measured before the PAM delivery point)

Position	May 2002		June 2002	
	Concentration (ppb)	Removal (%)	Concentration (ppb)	Removal (%)
Before PAM	10.56	-	3.18	-
Pond	1.41	-86.7	0.93	-70.7
104 m <sup>†</sup>	9.27	-12.2	1.11	-65.0
166 m	4.26	-59.6	0.55	-82.8
187 m	2.83	-73.2	0.43	-86.6
210 m	1.68	-84.1	0.95	-70.2
240 m <sup>‡</sup>	0.87	-91.8	0.28	-91.3
340 m	0.96	-90.9	0.30	-90.7

<sup>†</sup>Beginning of vegetative strip.

<sup>‡</sup>End of vegetative strip.





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## **The Ants Underground: Youth CD-ROM and Lesson Plans**

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### **Abstract**

Youth education is important and is a part of the fire ant education program. An interactive project aimed at educating youth aged 9 - 12 has been developed. Fire ant history, biology, and management is presented to youth using animation and sound.

### **Introduction**

Our youth fire ant education program has three major objectives: for youth to develop an understanding of basic fire ant biology, for youth to develop an understanding of proper management techniques, and the consequences of use of improper methods, and for adults to be impacted by youth, i.e., teaching youth may alter behavior of parents, especially in the common use of gasoline (Blanchard, 2000)

While one-time visits to a classroom are successful in introducing the youth to fire ants, a more in-depth presentation of fire ant information was needed to have an impact and change youth's attitude toward fire ants. Experience in the classroom with other educational materials (Project Wet, Project Wild) has shown that reinforcement of ideas increases the level of learning. Therefore, in June 1997 a Cooperative Extension Service workgroup discussed the development of the youth program using computer technology.

### **Production and Evaluation Stage**

Fire ant education was the purpose of the youth cd-rom, However, we wanted the program to be interactive. The interaction was to include self-selection of content view and opportunities to answer questions via computer. Areas of emphasis were to be the history, biology, and management of the red imported fire ant.

The storyboards were developed and clay models also. Lightwave (Newtek) and Poser (Curios Labs) were the 3D animation tools. Flash 4 was used for authoring, programming and interface design. Flash 4 and AfterEffects (Adobe) were used for 2D animation. Evaluation among Extension personnel has been a part of the program since its inception. Children of CES personnel have been asked to critique the material, and Pulaski Co. 4-H



groups have been involved on a formal basis. The initial evaluation by Arkansas 4-H'ers helped us in identifying problem language areas and the suitability of graphics. An evaluation of the games segment was conducted in Fall 2000 and Winter 2003. McPeake & Ballard (2000) videotaped the youth volunteers playing the games in Fall 2002, and a written evaluation was prepared. Based on the results of 2000 and 2003 there were several suggestions for improvement in the games section of the program.

## **Production Completed**

“The Ants Underground” provides an alternative to the traditional teaching methods that may reach a child not responsive to traditional teaching methods. Hart (1983) cited that a wide range of teaching techniques directly enhances the learning process and “The Ants Underground”, provides a wide-range of possibilities for its use. “The Ants Underground” story is told from the perspective of a native ant in the general debriefing section, and continues through the history, biology, and management. The program concludes with the participant receiving a clearance badge to go out and develop a plan of action for their community.

We have developed teacher lesson plans that tie into the current scientific education standards as part of “The Ants Underground” package. This tie-in is critical to reach a larger audience with our educational effort, by assuring the project's use by educators.

We have been evaluating the project from the storyboard stage and now are ready for its true field evaluation with teachers and other educators. After several revisions “The Ants Underground” is completed and will be distributed throughout Arkansas. We are in the process of developing training workshops for agents and teachers to assist in the successful use of the program.

Development of “The Ants Underground” has been very challenging. The rapidly changing technology has impacted the completion deadline. Use of the technology, and the financial resources involved in the production of the project were major concerns for the project.

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## **Cindy Heights Neighborhood Abatement Program Jefferson County, Arkansas**

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### **Introduction**

Neighborhood abatement programs are difficult to initiate in Arkansas. Communities have to take ownership in the program for the program to continue. In 2002, an African-American neighborhood began the first stage of developing a program. Gaining confidence in the Extension-recommended Two-Step method of fire ant management was the first step. Demonstrating the program's effectiveness and the neighborhood's own ability to minimize the presence of the red imported fire ant by community involvement was the second step.

The red imported fire ant is well established in Jefferson County, the county having been under the USDA/APHIS Quarantine since 1990. Pine Bluff with a population of 55,085 is the largest community in Jefferson Co. (population 84,278). The Cindy Heights Subdivision is located in the central area of the city, surrounded by schools and other subdivisions. It is not an isolated area.

### **Materials and Methods**

Community fire ant abatement programs are a part of many communities in Arkansas. Unlike demonstrations or educational programs where state Extension faculty or county Extension agents are responsible for program implementation, the community itself takes ownership of the program with assistance from Extension. Members of the Cindy Heights Neighborhood initiated a request to the Jefferson Co. Extension office for an abatement program in the summer 2002. Extension personnel became involved in trying to assist them with this request.

Fire ants were managed using the Extension-recommended Two-Step method. In late summer August 2002 the 68 house neighborhood was treated by broadcasting a bait product throughout the neighborhood. A contact product was provided to street captains to treat individual mounds on an on-request basis.

A house-to-house survey was conducted December 5, 2002 to assess individuals in the neighborhood's opinion about the program. Our goal was to assess program participation and effectiveness in changing perceptions about fire ant abatement and control methods.



These face-to-face interviews were conducted during daylight hours by visiting the respondents at home. Home addresses of respondents were mapped.

The interviewers worked together and questioned adult household members > 17 years old. After a brief introduction explaining the interviewers' affiliation and purpose of the survey, one interviewer asked the questions and another recorded responses. Questions were used to elicit a wide-range of responses about the program, its successes, and its failures. These questions were: 1: What did you think of the treatment program? 2: Did you see any changes in fire activity? 3: What were you doing to manage your fire ants previously? 4: Do you think your neighborhood will continue the program? Why or Why Not? 5: Would you be willing to pay for the treatment? If so, How Much?, and finally -- Any comments about the program you would like to make?

## **Results and Discussion**

One-on-one personal interviews were conducted by three interviewers on December 5 2002. Nineteen homes (28%) of the 67 homes in the subdivision were involved in the survey. Nineteen interviews were initiated, however only 17 were completed. Responses were recorded on paper and compiled. The average interview length was less than 5 minutes.

In this small neighborhood, the interviewers and respondents were familiar with one another prior to the interview. One interviewer was a street captain and had been involved in the abatement program from its initiation; the other interviewers were the local county extension agent and an extension specialist. There was no need to recruit a minority interviewer in that two of the three interviewees were African-American males. It is certain that their presence increased the response rates and openness of minority respondents (Tashakkori & Teddlie 1998). Studies show that the race of an interviewer can play a role in response bias, particularly if a standardized measurement process is not followed (Fowler & Mangione 1990). Weiss (1968) found in a study of welfare mothers that a higher rapport with the interviewer resulted in biased data, and this may have brought a certain bias to the survey. Because interviewers were known by respondents prior to the interviews, the interviewers stressed the need to know the good and bad about the program throughout the interview (Tashakkori & Teddlie 1998). A visual inspection indicated all block areas in the abatement program were sampled. The sample was deemed satisfactory. There were 9 male respondents and 8 female.

The following is a general overview of responses:

### **What did you think of the treatment program?**

Good, Fine – 15

Neutral – 2

Negative – 0

### **Did you see any changes in fire ant activity?**

Yes – 15

No - 0



Do not know – 2

**What were you doing to manage your fire ants previously?**

Nothing – 2

Unknown contact insecticide – 10

Pest Control Operator – 2

Gasoline – 1

Hot Water – 1

Amdro - 1

**Do you think the neighborhood will continue the program? Why or Why Not?**

Yes, I would like to see it continue – 16

Check with my husband - 1

**Would you be willing to pay for the treatment? If so, How Much?**

*Captain Johnny Jones asked if it was \$20.00 for a spring and fall treatment would they be willing to pay that amount.*

Yes– 17

Typical replies (I would be willing, I can afford it, and you are spending that much yourself)

**Any Comments about the program you would like to make?**

Glad to see the Ag program at work

Could we extend it to surrounding areas

I think it was nice the ants left

I didn't know what was going on, but noticed a decrease in fire ants

Glad to see someone take an interest in this neighborhood

That's good. It needs it – suckers were getting out of hand

**CONCLUSION**

Review of the responses show that the majority of the residents were satisfied with the results of the treatment. However, many were still unaware of the two-step program. From the terminology used by the respondents many have not taken ownership in the program in that they consistently stated 'I hope THEY will continue the program'. Further meetings are planned for 2003 to educate the neighborhood more extensively in the two-step fire ant management program and what their neighborhood can do to minimize fire ants.

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# Evaluation of broadcast applications of various contact insecticides against red imported fire ants, *Solenopsis invicta* Buren

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## Abstract

The efficacy of permethrin, bifenthrin, deltamethrin and fipronil was compared to that of various rates and formulations of imidacloprid and cyfluthrin against *Solenopsis invicta* Buren, red imported fire ants (RIFA). Efficacy was determined by collecting foraging RIFAs from each plot. Estimated percent reduction was 90, 66, and 45 for granular imidacloprid, granular cyfluthrin and the 9.55 l/ha (130.7 oz./acre) liquid cyfluthrin/imidacloprid formulation, at 1 DAT, respectively. By 65 days after treatment (DAT) only the granular fipronil treated plots had statistically significant ( $\alpha = 0.05$ ) fewer fire ants than the untreated control.

(Full article submitted to special issue of Journal of Agricultural and Urban Entomology)



**Biological Control Releases in Arkansas 2002:  
*Pseudacteon tricusps* and *Thelohania solenopsae***

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## Abstract

The phorid fly, *Pseudacteon tricusps*, and the microsporidia *Thelohania solenopsae* were released in Arkansas in 2002. Approximately 2885 and 1330 phorid flies were released in Pike and Bradley Counties, respectively. Initial results from Pike Co. suggest that the phorids have reproduced and expanded out of the immediate area of the release. Twenty-five fire ant mounds in Miller Co. were inoculated with *Thelohania solenopsae* infected brood.

## Introduction

Classical biological control attempts to reunite predators or parasites with their prey or host. Arkansas is attempting to reunite the phorid fly *Pseudacteon tricusps*, and the microsporidia *Thelohania solenopsae* with its natural host the red imported fire ant *Solenopsis invicta*.

*Pseudacteon tricusps* is a decapitating fly. The egg is oviposited by the female into the host and maggot's development occurs in the head of the fly. In the process of developing an enzyme is released by the fly which causes the tissues of the thorax to dissolve and the head falls off. However, it is not this action that makes the fly an effective biological control organism, it is the behavior the ants' exhibit when the flies are present – they hide, and are not efficient foragers.

*Thelohania solenopsae* is the most common fire ant pathogen in Brazil. It was discovered in the US in 1998 (FL, TX, MS, and OK). The microsporidia are obligate intracellular parasites which impact the longevity of the ant, the result is decreased colony size and colony density.

## *Pseudacteon tricusps* Release Method



# Evaluation of broadcast applications of various contact insecticides against red imported fire ants, *Solenopsis invicta* Buren

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## ***Pseudacteon tricusps* Release Method**



Using protocols developed at USDA-ARS Gainesville, Florida, the phorid fly, *Psuedacteon tricuspidis*, was released in Pike and Bradley counties in Arkansas. The sites were evaluated prior to the release for the presence of various habitat characteristics. Characteristics desired included a high population of fire ants – preferably monogyne colonies. The topography of the area selected should be diverse – including changes in elevation, diverse plant material from weeds to trees, and a water source. Both the release and control sites met these criteria.

Fly pupae were shipped overnight from USDA-ARS Florida and placed immediately in an emergence chamber. At approximately 11:00 am CDT on release days, the emergence chamber was placed in the aspirating chamber and approximately 30 flies were aspirated into each transport vial. All emerged flies in the aspirating chamber were collected into the transport vials in this manner until all flies were collected. At approximately 1:00 pm CDT the flies were released into attack chambers or actual mounds. Observations were made at 5 minute intervals and the numbers of flies observed were noted over a two hour period. The fly pupae were shipped May 9 and May 16, 2002 and the emerged flies released May 13 – May 26, 2002 in Pike Co. In Bradley Co. fly pupae were shipped October 1, 2002, and October 7, 2002, the emerged flies released October 3 – October 12, 2002.

### *Thelohania solenopsae* Release Method

Using protocols developed at USDA-ARS Gainesville, Florida the microsporidia *Thelohania solenopsae* was released in Miller Co. Arkansas.

The site was selected for the number of fire ant mounds, and the absence of the microsporidia. Samples were taken from the site in April 2002 and evaluated for the presence of the organism. Pools of 5-20 workers are prepared and observed with a phase contrast microscope for spores and *T. solenopsae* spores were not found.

Brood was shipped overnight from USDA-ARS Florida September 9, 2002. On Sept 11, 2002 mounds were evaluated and GPS coordinates of the mounds were taken (Fig 1, Fig 2). On Sept 12th, 1 - 2.5 grams of infected brood were placed into each of 25 mounds within a circular 1/16 acre plot.

### Results and Discussion

In 1998, releases of both *P. tricuspidis* and *T. solenopsae* were made; however, neither of the organisms is currently detectable. In 2002, releases of the organisms were attempted again. Initial data collection suggests that one of the organisms, *P. tricuspidis*, may be established in one location in Arkansas. Monitoring for establishment and impact of both organisms will resume in Spring 2003.

A slight difference in the percent emergence of *P. tricuspidis* pupal shipments for the two releases were noted (Fig 3). Fifty-two percent of the flies shipped for the May release emerged compared to 46% emergence from the October shipment. Knowledge of the



potential viability of the flies may impact future release periods, so as to maximize the potential for success.

The Pike Co. release site shows that for the spring and summer of 2002 the flies did reproduce (Fig 4) and were present outside of the release area. About 75% of the mounds 50 yards outside and in each direction (north, south, east and west) from the initial release area were positive for *P. tricuspidis* during the final evaluation of 2002. However, it is uncertain if the flies survived the winter of 2002. It is also uncertain if progeny from the flies released in Bradley Co. survived the winter. Regarding the observation on September 11, 2002 (Fig 4), it should be noted the observations were made approximately 10 hours after sunrise. Pesquero et al (1996) observed that *P. tricuspidis* exhibits a pattern of activity that peaks 7 to 9 hours after sunrise.

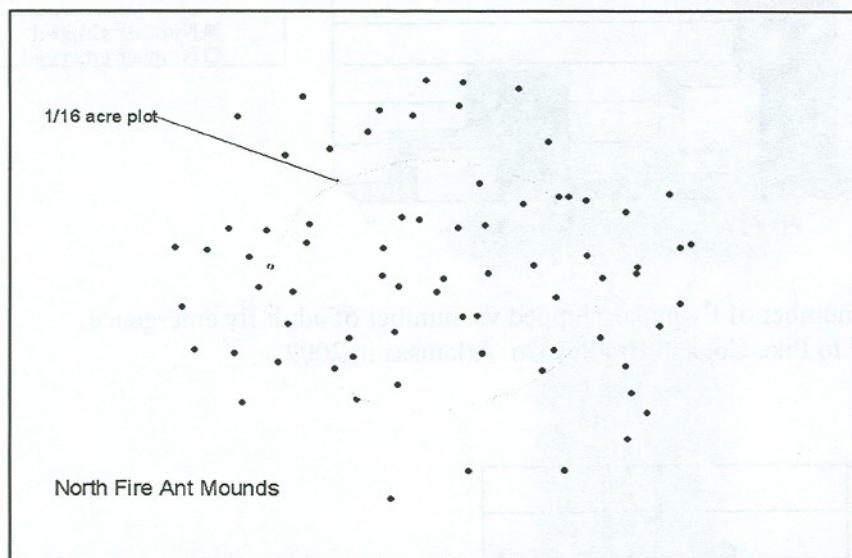
*T. solenopsae* was previously released in 1998 near Hope, Arkansas. Although *T. solenopsae* presence was noted following the 1998 release, recent surveys have not detected the pathogen. The 2002 release site, while similar in fire ant mound density and queen status to the 1998 release site, has a slightly different soil type which may benefit the disease organism.

Valuable experience was gained through the interagency cooperation on this project. Participants learned a great deal regarding techniques and potential factors to look for in the success of any future releases. In addition these releases provided valuable outreach opportunities for county agents associated with this project.

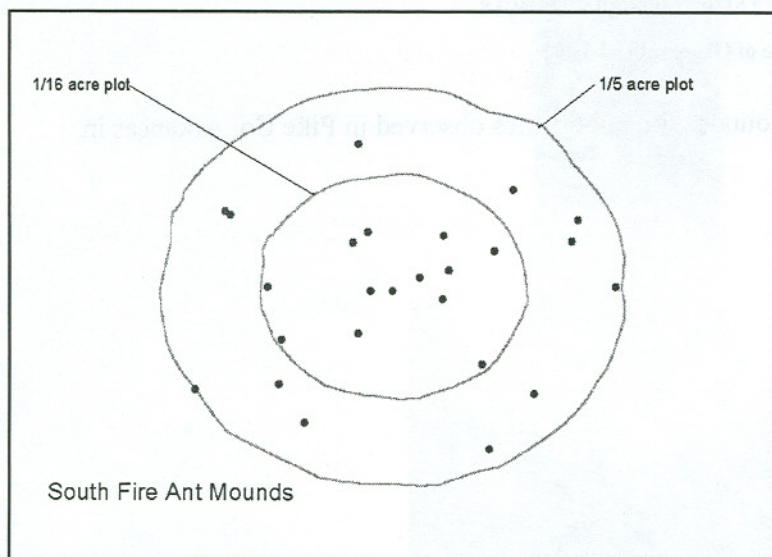
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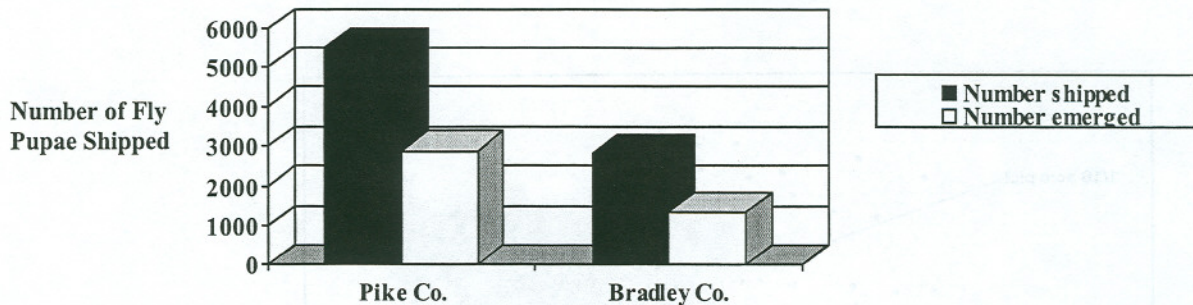


**Fig 1.** Location of fire ant mounds within a 1/5 acre plot in Miller Co., Arkansas.  
*T. solenopsae* released within the circle

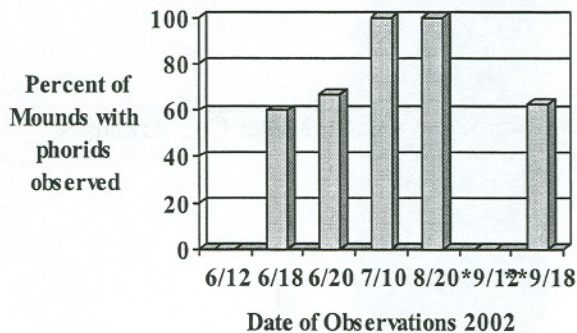


**Fig 2.** Location of fire ant mounds within a 1/5 acre plot in Miller Co. Arkansas,  
 Control site





**Fig 3.** Comparison of the number of fly pupae shipped vs. number of adult fly emergence from the fly pupae shipped to Pike Co. and Bradley Co. Arkansas in 2002.



**Fig 4.** Percentage of evaluated mounds with phorid flies observed in Pike Co. Arkansas in 2002.



**Morphological Embryonic Development of the Red Imported Fire Ant *Solenopsis invicta* Buren (Hymenoptera: Formicidae)**

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**Abstract**

Embryonic characteristics during embryonic development was studied by external observation of the red imported fire ant *Solenopsis invicta*. Early embryonic patterning belonged to the long-germ-type. The germ band formed at the ventral side of the egg, composed of the incipient head lobes, the gnathal and thoracic region, and a growth zone. The germ band developed by extending and curving dorsally in both anterior and posterior directions, until the head and the tail almost connected with each other on the dorsal side of the egg when the germ band reached its maximum length. In the mean time, the segmentation and appendages appeared on the germ band. Five distinctive embryonic stages were evident according to the external characteristics of the growing embryos.



## Preliminary assessment of *Dorymyrmex* and *Solenopsis invicta* interactions

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### Introduction

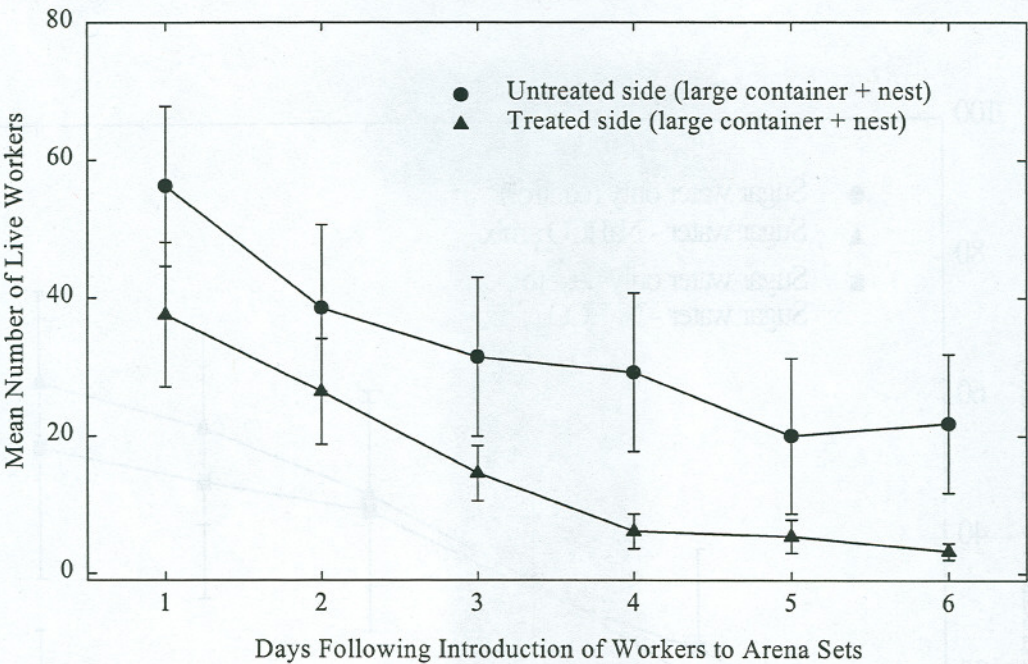
Hung (1974) made preliminary observations on fire ants (*Solenopsis invicta* Buren) recovered from a single refuse pile of the Pyramid ant *Conomyrma insana* (= *Dorymyrmex* Corel) and found fire ants predominated (43%); some authors (Smith 1965; Nickerson et al. 1975; Roe 1974; Wilson et al. 1971) suggested *Dorymyrmex* may kill newly mated queens, alate males and workers, and that perhaps it sometimes nests at the edge of the fire ant mounds, but these observations have not been quantified. The role of native ants in habitats treated for fire ants is not yet well understood. More knowledge of the interactions of fire ants and native ants is needed. We observed fire ant remains adjacent to enters of *Dorymyrmex flavus* McCook nests in a commercial pecan orchard in Burleson Co., TX while conducting extensive investigations on fire ant in pecan. The *Dorymyrmex* nests were located in plots previously treated with Extinguish® bait (applied on May 19, and October 12, 2000 and June 12 in 2001) and had fire ant nests nearby. Fire ant nest density had been reduced in these plots by about 70%. We documented the observations by collecting the midden (a raised, loosely aggregated mound of earth, sticks, dead ants, ant parts, and other materials constructed next to the nest entrance by *Dorymyrmex*) from each of ten nests on July 31, 2002 and again on Aug. 8, 2002. Middens were carefully inspected with the aid of a binocular microscope (4-60x) searching for arthropod remains and other materials that might be of interest. More than 98% of the arthropod remains found consisted of fire ants, with the initial inspection on July 31 showing an average minimum of 203 dead fire ants per midden and the Aug. 8 inspection showing 40 per midden. Further investigations are being conducted to determine how the fire ants came to be in the middens and whether or not these findings are important in fire ant management.

### Materials and Methods

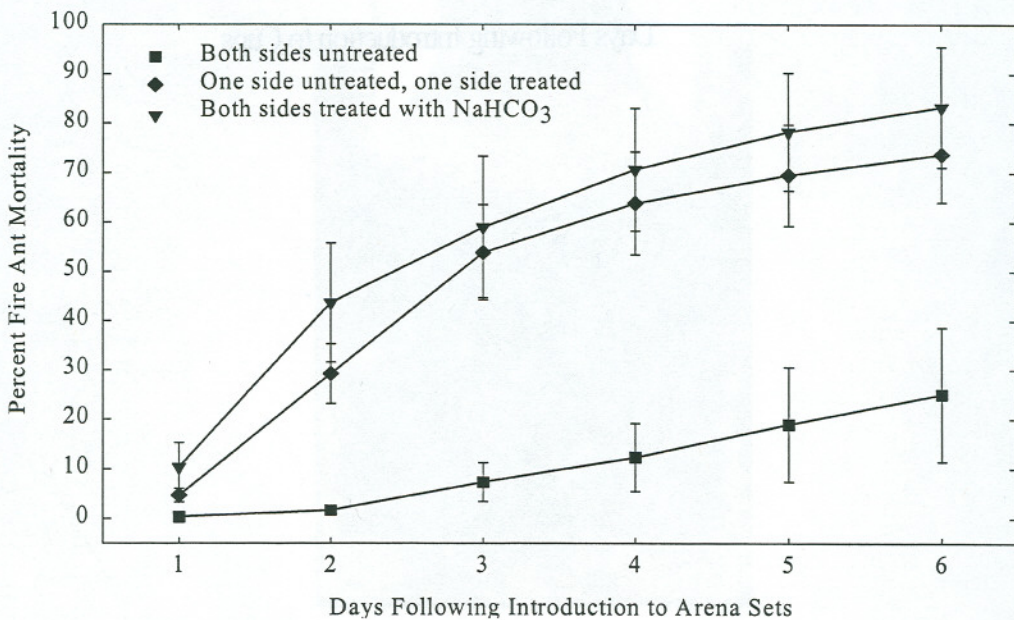
The study site is located in a commercial pecan orchard at Mumford, TX (Robertson Co. 30° 44' 55" N; 96° 33' 12" W). Ten middens of *Dorymyrmex* were collected randomly in an area previously treated for fire ants with methoprene bait Extinguish® (an Insect Growth Regulator (IGR) applied on May 19, and October 12, 2000 and June 12 in 2001). The entire midden was collected from each nest on July 31, 2002 and nests were marked and referenced with GPS; middens were again collected on August 8 2002 from the same nests. The middens were placed in plastic snap cap vials (7 dram) and returned to the lab and cleaned of debris and soil particles, and all insect remains were sorted and preserved in 70% alcohol for further inspection. Each sample was initially divided into a 1.5 ml subsample by volume. Then each subsample was inspected and sorted. Arthropods and body parts were placed in vials with 70% alcohol. Ants were identified and counted, and remains of the body classified as: body= if body is found complete with the head, thorax and abdomen; head= if only head is visible; thorax= if only thorax is visible; abdomen= if only abdomen is visible; head + thorax= if only head and thorax is visible; thorax + abdomen= if only thorax and



**Fig. 1.** Mean number of live *S. invicta* in untreated sides of arena sets and in sides treated with  $\text{NaHCO}_3$ . Ants ( $n = 100$ ) were added to a smaller central container and could move freely between treated and untreated sides through vinyl tubing. Each side was comprised of a large container plus artificial nest.

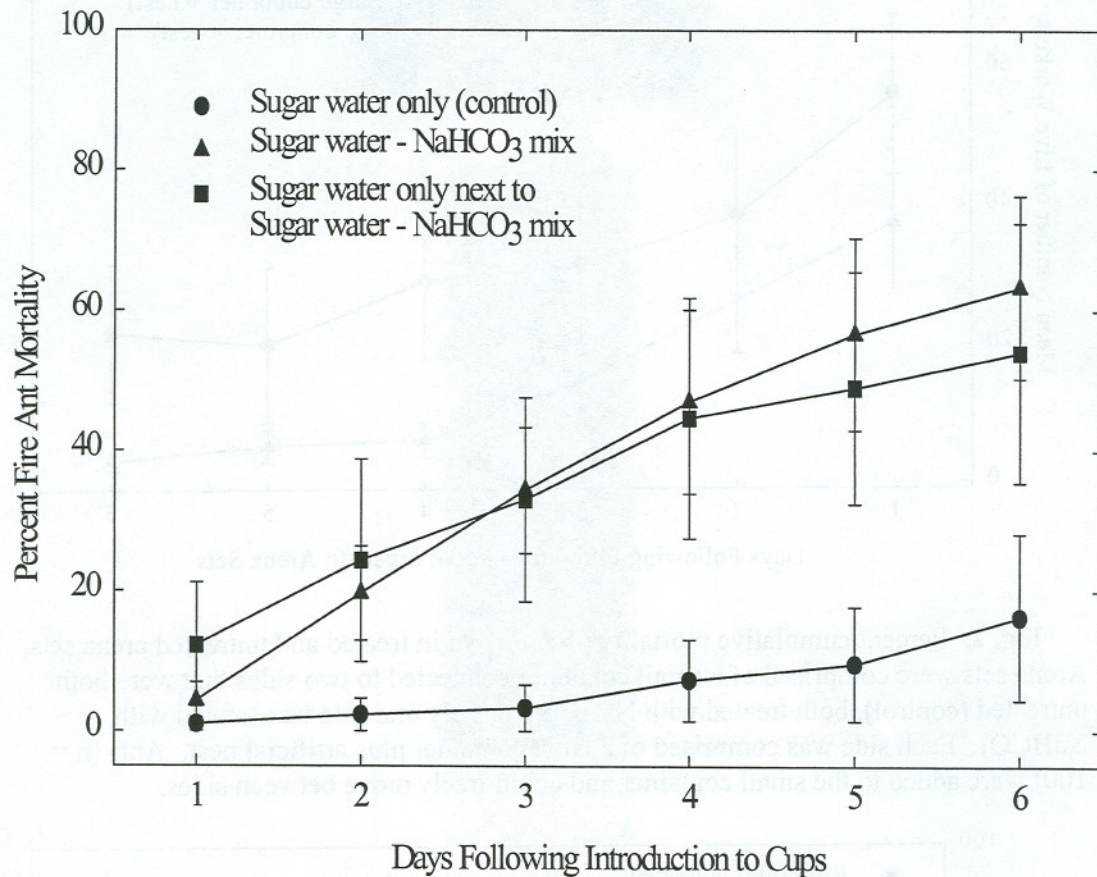


**Fig. 2.** Percent cumulative mortality of *S. invicta* in treated and untreated arena sets. Arena sets were comprised of a small container connected to two sides that were both untreated (control), both treated with  $\text{NaHCO}_3$ , or only one side was treated with  $\text{NaHCO}_3$ . Each side was comprised of a large container plus artificial nest. Ants ( $n = 100$ ) were added to the small container and could freely move between sides.





**Fig. 3.** Percent cumulative mortality of *S. invicta* provided untreated sugar water (control), NaHCO<sub>3</sub>-sugar water mix, or untreated sugar water in a container next to a container with NaHCO<sub>3</sub>-sugar water mix.





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*(Note: These “snapshots” are single page printouts of each poster. The original Poster files are on the CD provided with these Proceedings and are identified by the page numbers and first author, as listed below.)*

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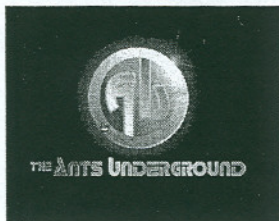
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# The Ants Underground: Youth CD-ROM and Lesson Plans

Donna R. Shanklin<sup>1</sup>, Kelly M. Loftin<sup>2</sup>, Tom Riley<sup>3</sup>,  
Chris Meux<sup>4</sup>, Jason Shivers<sup>4</sup>, and Ed Rhodes<sup>4</sup>, J. Brian Richardson<sup>5</sup>

University of Arkansas Cooperative Extension Service



Ants Underground Logo

## Abstract

Youth education is important and is a part of the fire ant education program. An interactive project aimed at educating youth aged 9 - 12 has been developed. Fire ant history, biology, and management is presented to youth using animation and sound.



Depiction of Red Imported Fire Ants

## Introduction

Our youth fire ant education program has three major objectives:

- For youth to develop an understanding of basic fire ant biology
- For youth to develop an understanding of proper management techniques, and the consequences of use of improper methods
- For adults to be impacted by youth, i.e., teaching youth may alter behavior of parents, especially in the common use of gasoline (Blanchard, 2000)

While one-time visits to a classroom are successful in introducing the youth to fire ants, a more in-depth presentation of fire ant information was needed to have an impact and change youth's attitude toward fire ants. Experience in the classroom with other educational materials (Project Wet, Project Wild) has shown that reinforcement of ideas increases the level of learning. Therefore, in June 1997 a Cooperative Extension Service workshop discussed the development of the youth program using computer technology

## Production and Evaluation Stage

Fire ant education was the purpose of the youth cd-rom. However, we wanted the program to be interactive. Interaction to include selection of content view and opportunities to answer questions via computer. Areas of emphasis were to be history, biology, and management of the red imported fire ant.

The storyboards were developed (Fig 1) and clay models (Fig 2). Lightwave (Newtek) and Poser (Curious Labs) were the 3D animation tools. Flash 4 was used for authoring, programming and interface design. Flash 4 and AfterEffects (Adobe) were used for 2D animation. Evaluation among Extension personnel has been a part of the program since its inception. Children of CES personnel have been asked to critique the material, and Pulaski Co. 4-H groups have been involved on a formal basis. The initial evaluation by Arkansas 4-Hers helped us in identifying problem language areas and the suitability of graphics. An evaluation of the games segment was conducted in Fall 2000 and Winter 2003. McPeake & Ballard (2000) videotaped the youth volunteers playing the games in Fall 2002, and a written evaluation was prepared. Based on the results of 2000 and 2003 there were several suggestions for improvement in the games section of the program.

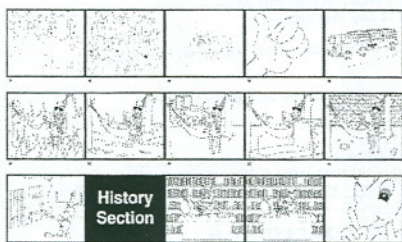


Fig 1. Storyboard



Fig 2. Clay model of ant figure

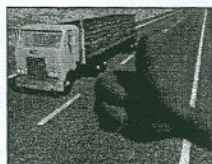


Fig 3. Image from General Debriefing

## Production Completed

"The Ants Underground" provides an alternative to the traditional teaching methods that may reach a child not responsive to traditional teaching methods. Hart (1983) cited that a wide range of teaching techniques directly enhances the learning process and we feel "The Ants Underground", provides a wide-range of possibilities for its use. "The Ants Underground" story is told from the perspective of a native ant in the general debriefing section, and continues to the history, biology, and management. The program concludes with the participant receiving a clearance badge to go out develop a plan of action for their community.

We have developed teacher lesson plans that tie into the current scientific education standards as part of "The Ants Underground" package. This tie-in is critical to reach a larger audience with our educational effort, by assuring the project's use by educators.

We have been evaluating the project from the storyboard stage and now are ready for its true field evaluation with teachers and other educators. After several revisions "The Ants Underground" is completed and will be distributed throughout Arkansas. We are in the process of developing training workshops for agents and teachers to assist in the successful use of the program.

Development of "The Ants Underground" has been very challenging. The rapidly changing technology has impacted the completion deadline. Use of the technology, and the financial resources involved in the production of the project were major concerns for the project.

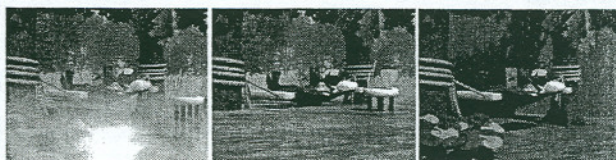


Fig 4. Sequence of images from General Debriefing

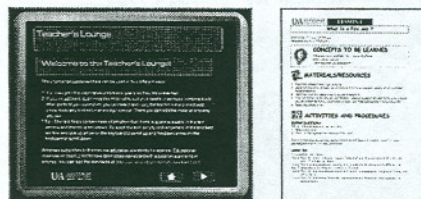


Fig 5. Teacher Lounge Page and PDF version of Lesson 1

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# Cindy Heights, Neighborhood Abatement Program, Jefferson County, Arkansas

Joe Ivy<sup>1</sup>, Donna Shanklin<sup>2</sup>, Kelly Loftin<sup>3</sup> and John Hopkins<sup>3</sup>

University of Arkansas Cooperative Extension Service



## Introduction

Neighborhood abatement programs are difficult to initiate in Arkansas. Communities have to take ownership in the program for the program to continue. In 2002, an African-American neighborhood began the first step of developing a program. Gaining confidence in the Extension-recommended Two-Step method of fire ant management was the first step. Demonstrating the program's effectiveness and the neighborhood's own ability to minimize the presence of the red imported fire ant by community involvement was the second step.

The red imported fire ant is well established in Jefferson County, the county having been under the USDA/APHIS Quarantine since 1990. Pine Bluff with a population of 55,085 is the largest community in Jefferson Co. (population 84,278). The Cindy Heights Subdivision is located in the central area of the city, surrounded by schools and other subdivisions (Fig 4). It is not an isolated area.

## Materials and Methods

Community fire ant abatement programs are a part of many communities in Arkansas. Unlike demonstrations or educational programs where state Extension faculty or county Extension agents are responsible for program implementation, the community itself takes ownership of the program with assistance from Extension. Members of the Cindy Heights Neighborhood initiated a request to the Jefferson Co. Extension office for an abatement program in the summer 2002. Extension personnel became involved in trying to assist them with this request.

Fire ants were managed using the Extension-recommended Two-Step method. In late summer August 2002 the 68 house neighborhood was treated by broadcasting a bait product throughout the neighborhood (Fig 1, and Fig 2) A contact product was provided to street captains to treat individual mounds on an on-request basis.

A house-to-house survey was conducted December 5, 2002 to assess individuals in the neighborhood's opinion about the program. Our goal was to assess program participation and effectiveness in changing perceptions about fire ant abatement and control methods. These face-to-face interviews were conducted during daylight hours by visiting the respondents at home (Fig 3). Home addresses of respondents were mapped (Fig 4).

The interviewers worked together and questioned adult household members > 17 years old. After a brief introduction explaining the interviewers' affiliation and purpose of the survey, one interviewer asked and another recorded responses to five questions. Questions were used to elicit a wide-range of responses about the program, its successes, and its failures. These questions were: 1: What did you think of the treatment program? 2: Did you think it had an impact? 3: What were you using prior to the treatment? 4: Would you be willing to continue the program and if so. 5: Any comments you would like to make

## Results and Discussion

One-on-one personal interviews were conducted by three interviewers on December 5 2002. Nineteen homes (28%) of the 67 homes in the subdivision were involved in the survey. Nineteen interviews were initiated, however only 17 were completed. Responses were recorded on paper and compiled. The average interview length was less than 5 minutes.

In this small neighborhood, the interviewers and respondents were familiar with one another prior to the interview. One interviewer was a street captain and had been involved in the abatement program from its initiation, the other interviewers were the local county extension agent and an extension specialist. There was no need to recruit a minority interviewer in that two of the three interviewers were African-American males. It is certain that their presence increased the response rates and openness of minority respondents (Tashakkori & Teddlie 1998). Studies show that the race of an interviewer can play a role in response bias, particularly if a standardized measurement process is not followed (Fowler & Mangione 1990). Weiss (1968) found in a study of welfare mothers that a higher rapport with the interviewer resulted in biased data, and this may have brought a certain bias to the survey. Because interviewers were known by respondents prior to the interviews, the interviewers stressed the need to know the good and bad about the program throughout the interview (Tashakkori & Teddlie 1998). A visual inspection indicated all block areas in the abatement program were sampled (Figure 4). The sample was deemed satisfactory. There were 9 male respondents and 8 female.

The following is a general overview of responses:

**What did you think of the treatment program?**

Good, Fine - 15  
Neutral - 2  
Negative - 0

**Did you see any changes in fire ant activity?**

Yes - 15  
No - 0  
Do not know - 2

**What were you doing to manage your fire ants previously?**

Nothing - 2  
Unknown contact insecticide - 10  
Pest Control Operator - 2  
Gasoline - 1  
Hot Water - 1  
Amdro - 1

**Do you think the neighborhood will continue the program? Why or Why Not?**

Yes, I would like to see it continue - 16  
Check with my husband - 1

**Would you be willing to pay for the treatment? If so, How Much?**

Captain Johnny Jones asked if it was \$20.00 for a spring and fall treatment would they be willing to pay that amount.

Yes - 17

Typical replies (I would be willing, I can afford, and you are spending that much yourself)

**Any Comments about the program you would like to make?**

Glad to see the Ag program at work  
Could we extend it to surrounding areas  
I think it was nice the ants left  
I didn't know what was going on, but noticed a decrease in fire ants  
Glad to see someone take an interest in this neighborhood  
That's good. It needs it - suckers were getting out of hand

## CONCLUSION

Review of the responses show that the majority of the residents were satisfied with the results of the treatment. However, many were still unaware of the two-step program. From the terminology used by the respondents many have not taken ownership in the program in that they consistently stated 'I hope THEY will continue the program'. Further meetings are planned for 2003 to educate the neighborhood more extensively in the two-step fire ant management program and what their neighborhood can do to minimize fire ants.

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Fig 1. Street Captains and CES agent Ivy Prepare to treat neighborhood



Fig 2. CES Agent Ivy Evaluates effectiveness

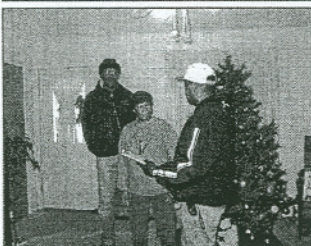


Fig 3. Street Captain Johnny Jones and CES agent Ivy questioning Emma Jones, neighborhood member

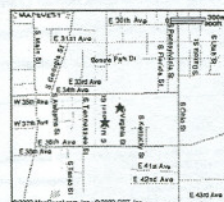


Fig 4. Cindy Heights Neighborhood



# Evaluation of broadcast applications of various contact insecticides

John Gavin<sup>1</sup>, Kelly Loftin<sup>2</sup>, Donna Shanklin<sup>3</sup>, and John Hopkins<sup>2</sup>

UNIVERSITY OF ARKANSAS  
DIVISION OF AGRICULTURE

University of Arkansas Cooperative Extension Service

## Abstract

The objective of this trial was to evaluate the efficacy of broadcast applications of 0.0103% fipronil granules, 0.1% cyfluthrin granules, 0.2% imidacloprid granules, and two rates of cyfluthrin/imidacloprid liquid against red imported fire ants, *Solenopsis invicta*. At one day post-treatment the imidacloprid granular, cyfluthrin/imidacloprid 3 oz/acre liquid, and cyfluthrin granular treatments had statistically significant fewer fire ants than the untreated control. The estimated percent reduction was 90, 66, and 45 for imidacloprid granular, cyfluthrin granular and cyfluthrin/imidacloprid 3 oz/acre liquid, at day 1 post treatment, respectively. The imidacloprid granular treatment maintained control through 7 days. The bifenthrin and fipronil granular treatments showed significant reduction at 3 days. Fipronil treatment was the only insecticide treatment to show a statistically significant reduction in fire ants and the proportion of mounds containing brood (egg, larva, pupa). This amounted to a 56% reduction when compared to the untreated control.

## Materials and Methods

**Study Site:** The study was conducted in Rye, Arkansas located in Cleveland County in southeast Arkansas. Density of red imported fire ant, *Solenopsis invicta* mounds at the study site was approximately 240 mounds per acre. Grass had been mowed approximately 3 days prior to the study.

**Design:** Each treatment and the untreated control was replicated in 3 plots. Plots were approximately 0.25 acres in size. The treatments and control were randomly allocated within each block. The three blocks corresponded to different areas of the study site.

**Evaluation:** Pre and post treatment evaluations were conducted using bait stations to collect foraging ants within each plot. Bait stations consisted of a 1/4 inch hot dog cube placed on a snap vial lid and marked with a wire survey flag. Ten bait stations (two transects with 5 bait stations each, located in the center of each plot, transects were approximately 30 feet apart, bait stations within each transect were approximately 30 feet apart) in each plot were made available to foraging ants for approximately 30 minutes. The numbers of red imported fire ants estimated at each station in each plot was used to evaluate product efficacy. Percentage of mounds containing brood was determined during the final evaluation. Ten mounds in each plot were excavated and observed for the presence of fire ant brood (eggs, larva and pupa).

**Insecticide Applications:** Products and application rates are given in Table 1. Insecticides were broadcast using a Herd® seeder mounted on a Kawasaki® mule equipped with a digital speedometer. After calibration, the specified rates were broadcast by matching to the appropriate ground speed and dispersal rate. Liquid insecticide applications were made with a trailer mounted boom sprayer pulled by a Kawasaki® mule equipped with a digital speedometer. After calibration of the boom sprayer, a test application outside of treated area and with water only was made to determine the speed and amount of water required for uniform coverage. Insecticide applications were made on August 6, 2002.

**Statistical Analysis:** Data collected from bait stations were analyzed using Analysis of Variance based on a RCB design with 3 replicates. The protected LSD procedure was used to determine significant differences in the mean number of foraging RIFA=s collected from the various treatments (Statistix 2000). The percentage of mounds containing brood was analyzed using the same procedures as above.

Table 1. Insecticide rates and manufacturer

Insecticide	Rate	Manufacturer
0.25% permethrin GR	87lb/acre	Real Kill Multi Purpose
0.147% bifenthrin GR	136lb/acre	Scotts MaxGuard Insect Protection with Turf Builder
0.2% imidacloprid GR	65lb/acre	Bayer Advanced Lawn Grub Control
0.0103% fipronil GR	87 lb/acre	Over N Out
0.1% deltamethrin GR	87 lb/acre	Delta Shield
0.72% cyfluthrin EW and 0.72% imidacloprid	43.5oz/acre	Research stock supplied by Bayer-Purcell LLC.
0.72% cyfluthrin EW and 0.72% imidacloprid	130.7oz/acre	Research stock supplied by Bayer-Purcell LLC.
0.1% cyfluthrin GR	130lb/acre	Bayer Advanced Power Force



## Results and Discussion

The mean number of foraging fire ants estimated at bait stations is given in Table 2. At one day post-treatment the imidacloprid granular, cyfluthrin/imidacloprid 3 oz/acre liquid, and cyfluthrin granular treatments had statistically significant fewer fire ants than the untreated control. The estimated percent reduction was 90, 66, and 45 for imidacloprid granular, cyfluthrin granular and cyfluthrin/imidacloprid 3 oz/acre liquid, at day 1 post treatment, respectively (Table 3). The imidacloprid granular treatment maintained this trend through 7 days post treatment. The bifenthrin and fipronil granular treatments began showing significant reduction in the number of ants collected from bait stations at 3 days post treatment. The fipronil granular treatment maintained significant levels of control only through 14 days post treatment. Significant reductions in the number of ants observed at bait stations were never achieved by deltamethrin, permethrin or the cyfluthrin/imidacloprid 1 oz/acre combination. By 65 days post treatment only the fipronil plots had statistically significantly fewer fire ants than the control.

The mean percentage of fire ant mounds with brood is given in Table 2. Fipronil treatment was the only insecticide treatment to show a statistically significant reduction in the proportion of mounds containing brood (egg, larva, pupa). This amounted to a 56% reduction when compared to the untreated control (Table 3).

Table 2. Mean number of red imported fire ants per bait station. Brood (far right column) is the percentage of fire ant mounds containing brood (eggs, larva, pupa).

Insecticide	DAYS							
	0	1	2	3	7	14	35	65
Untreated Control	32.5	50.8 <sup>a</sup>	36.5 <sup>a</sup>	22.2 <sup>ab</sup>	71.7 <sup>ab</sup>	42.8 <sup>ab</sup>	55.8 <sup>ab</sup>	56.8 <sup>ab</sup>
0.25 % Permethrin granule	52.0	52.5 <sup>a</sup>	38.3 <sup>a</sup>	33.5 <sup>a</sup>	70.0 <sup>ab</sup>	50.0 <sup>ab</sup>	57.5 <sup>a</sup>	70.8 <sup>a</sup>
0.147% bifenthrin granule	34.0	34.2 <sup>abc</sup>	30.0 <sup>a</sup>	15.8 <sup>abc</sup>	34.6 <sup>ab</sup>	18.7 <sup>cd</sup>	42.0 <sup>ab</sup>	37.0 <sup>bc</sup>
0.2% imidacloprid granule	49.2	5.2 <sup>d</sup>	7.5 <sup>b</sup>	4.2 <sup>d</sup>	19.8 <sup>a</sup>	37.7 <sup>abc</sup>	36.7 <sup>b</sup>	58.5 <sup>ab</sup>
0.0103 fipronil granule	37.5	49.2 <sup>ab</sup>	27.8 <sup>a</sup>	9.5 <sup>cd</sup>	43.3 <sup>abc</sup>	13.5 <sup>d</sup>	9.50 <sup>d</sup>	16.0 <sup>d</sup>
0.1% deltamethrin granule	39.5	39.7 <sup>abc</sup>	31.1 <sup>a</sup>	20.8 <sup>ab</sup>	55.0 <sup>abc</sup>	35.0 <sup>abc</sup>	45.8 <sup>ab</sup>	61.2 <sup>ab</sup>
0.72% cyfluthrin + 0.72% imidacloprid spray (1.0oz/100 sq ft)	33.3	40.4 <sup>ab</sup>	34.2 <sup>a</sup>	23.3 <sup>ab</sup>	70.0 <sup>ab</sup>	54.2 <sup>a</sup>	48.3 <sup>ab</sup>	61.2 <sup>ab</sup>
0.72% cyfluthrin + 0.72% imidacloprid spray (3.0oz/100 sq ft)	40.8	27.8 <sup>cd</sup>	32.3 <sup>a</sup>	21.7 <sup>ab</sup>	75.8 <sup>a</sup>	52.0 <sup>a</sup>	60.1 <sup>a</sup>	60.0 <sup>ab</sup>
0.1% cyfluthrin granule	31.0	17.0 <sup>cd</sup>	21.3 <sup>ab</sup>	21.5 <sup>ab</sup>	50 <sup>abc</sup>	32.2 <sup>cd</sup>	46.0 <sup>ab</sup>	53.8 <sup>ab</sup>

Means followed by the same letter are not significantly different using the protected LSD mean separation procedure ( $\alpha = 0.05$ ).

Table 3. Estimated percent control of red imported fire ants after broadcast application with selected insecticides. Brood represents the estimated percent reduction in the proportion of mounds containing brood (eggs, larva, pupa).

Insecticide	DAYS							
	1	2	3	7	14	35	65	Brood
0.25 % permethrin granule	0	0	0	2	0	0	0	0
0.147% bifenthrin granule	33	18	29	51	56	25	33	5
0.2% imidacloprid granule	90	79	81	72	12	34	0	11
0.0103% fipronil granule	3	24	57	40	68	83	72	56
0.1% deltamethrin granule	22	15	6	23	18	18	0	0
0.72% cyfluthrin + 0.72% imidacloprid spray (1.0oz / 100 sq ft)	20	6	0	2	0	13	0	5
0.72% cyfluthrin + 0.72% imidacloprid spray (3.0oz/100 sq ft)	45	11	2	0	0	0	0	0
0.1% cyfluthrin granule	66	42	3	30	27	18	5	5

Means followed by the same letter are not significantly different using the protected LSD mean separation procedure ( $\alpha = 0.05$ ).

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Statistix 7. 2000. Analytical Software, Tallahassee, Florida

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# Biological Control Releases in Arkansas 2002: *Pseudacteon tricuspidis* and *Thelohania solenopsae*

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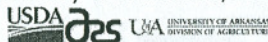


Fig 1. 2002 Release Sites in Arkansas  
- *Thelohania solenopsae*  
- *Pseudacteon tricuspidis*



Fig 2. Phorids mating



Fig 3. *Thelohania* spores



Fig 4. *Thelohania* cysts in worker ant

## Abstract

The phorid fly, *Pseudacteon tricuspidis*, and the microsporidia *Thelohania solenopsae* were released in Arkansas in 2002. Approximately 2885 and 1330 phorid flies were released in Pike and Bradley Counties, respectively. Initial results from Pike Co. suggest that the phorids have reproduced and expanded out of the immediate area of the release. Twenty-five fire ant mounds in Miller Co. were inoculated with *Thelohania solenopsae* infected brood.

## Introduction

Classical biological control attempts to reunite predators or parasites with their prey or host. Arkansas is attempting to reunite the phorid fly *Pseudacteon tricuspidis*, and the microsporidia *Thelohania solenopsae* with its natural host the red imported fire ant *Solenopsis invicta* (Fig 1).

*Pseudacteon tricuspidis*, is a decapitating fly. The egg is oviposited by the female into the host and maggot's development occurs in the head of the fly. In the process of developing an enzyme is released by the fly which causes the tissues of the thorax to dissolve and the head falls off. However, it is not this action that makes the fly an effective biological control organism, it is the behavior the ants exhibit when the flies are present – they hide, and are not efficient foragers (Fig 2).

*Thelohania solenopsae* is the most common fire ant pathogen in Brazil. It was discovered in the US in 1998 (FL, TX, MS, OK). The microsporidia are obligate intracellular parasites which impact the longevity of the ant (Fig 3 and Fig 4) The result is decreased colony size and colony density.

## *Pseudacteon tricuspidis* Release Method

Using protocols developed at USDA-ARS Gainesville, Florida the phorid fly, *Pseudacteon tricuspidis*, was released in Pike and Bradley County Arkansas.

The sites were evaluated prior to the release for the presence of various habitat characteristics. Characteristics including a high population of fire ants – preferably monogyne colonies. The topography of the area selected should be diverse – including changes in elevation, diverse plant material from weeds to trees, and a water source. (Fig 5 and Fig 6). Both the release and control sites met these criteria.

Fly pupae were shipped overnight from USDA-ARS Florida and placed immediately in an emergence chamber (Fig 7). At approximately 11:00 am CDT on release days, the emergence chamber was placed in the aspirating chamber and approximately 30 flies were aspirated into each transport vial (Fig 8 and Fig 9). All emerged flies in the aspirating chamber were collected into the transport vials in this manner until all flies were collected. At approximately 1:00 pm CDT the flies were released into attack chambers or actual mounds (Fig 10 and 11). Observations were made at 5 minute intervals and the number of flies observed were noted over a two hour period. The fly pupae were shipped May 9 and May 16, 2002 and the emerged flies released May 13 – May 26, 2002 in Pike County. In Bradley Co. fly pupae were shipped October 1, 2002, and October 7, 2002, the emerged flies released October 3 – October 12, 2002.

## *Thelohania solenopsae* Release Method

Using protocols developed at USDA-ARS Gainesville, Florida the microsporidia *Thelohania solenopsae* was released in Miller Co. Arkansas.

The site was selected for the number of fire ant mounds, and the absence of the microsporidia. Samples were taken from the site in April 2002 and evaluated for the presence of the organism. Pools of 5-20 workers are prepared and observed with a phase contrast microscope for spores and *T. solenopsae* spores were not found.

Brood was shipped overnight from USDA-ARS Florida September 9, 2002. On Sept 11, 2002 mounds were evaluated and GPS coordinates of the mounds were taken (Fig 12, Fig 13, Fig 14). On Sept 12<sup>th</sup>, 1-2.5 grams of infected brood were placed into each of 25 mounds within a circular 1/16 acre plot (Fig 15).

### References Cited

Pasquero, M. A., S. Camplido, H. G. Fowler, S. D. Porter, 1996, Diurnal patterns of ovipositional activity in two *Pseudacteon* fly parasitoids (Diptera: Phoridae) of *Solenopsis* fire ants (Hymenoptera: Formicidae), *In* Scientific Notes, Florida Entomologist 79(2), pp 455 – 457

## Results and Discussion

In 1998, releases of both *P. tricuspidis* and *T. solenopsae* were made, however, neither of the organisms is currently detectable. In 2002, releases of the organisms were attempted again. Initial data collection suggests that one of the organisms, *P. tricuspidis*, may be established in one location in Arkansas. Monitoring for establishment and impact of both organisms will resume this spring.

A slight difference in the percent emergence of *P. tricuspidis* pupal shipments for the two releases were noted (Fig 16). Fifty-two percent of the flies shipped for the May release emerged compared to 46% emergence from the October shipment. Knowledge of the potential viability of the flies may impact future release periods, so as to maximize the potential for success.

The Pike county release site shows that for the spring and summer of 2002 the flies did reproduce (Fig 17) and were present outside of the release area. About 75% of the mounds 50 yards outside and in each direction (north, south, east and west) from the initial release area were positive for *P. tricuspidis* during the final evaluation of 2002. However, it is uncertain if the flies survived the winter. It is also uncertain if progeny from the flies released in Bradley Co. survived the winter. Regarding the observation on September 11, 2002 (Fig 17), it should be noted the observations were made approximately 10 hours after sunrise. Pasquero et al (1996) observed that *P. tricuspidis* exhibits a pattern of activity that peaks 7 to 9 hours after sunrise.

*T. solenopsae* was previously released in 1998 near Hope, Arkansas. Although *T. solenopsae* presence was noted following the 1998 release, recent surveys have not detected the pathogen. The 2002 release site, while similar in fire ant mound density and queen status to the 1998 release site, contains a slightly different soil type.

Valuable experience was gained through the interagency cooperation on this project. Participants learned a great deal regarding techniques and potential factors to look for in the success of any future releases. In addition these releases provided valuable outreach opportunities for county agents associated with this project.

Fig 5. Pike Co. AR phorid release site



Fig 6. Bradley Co. phorid release site



Fig 7. Emergence chamber



Fig 8. Transport vial

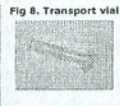


Fig 9. Aspirating chamber



Fig 10. Observing phorids released into attack chambers



Fig 11. Observing phorids released directly into a mound



Fig 12. Aerial View of *T. solenopsae* control and release sites

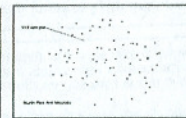


Fig 13. Map of mounds in *T. solenopsae* release site

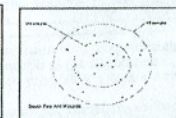


Fig 14. Map of mounds in *T. solenopsae* control site



Fig 15. OI and Loftin inoculating a mound with *T. solenopsae*

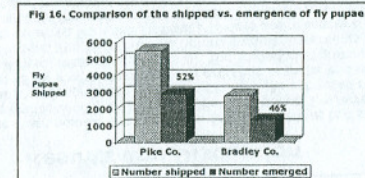


Fig 16. Comparison of the shipped vs. emergence of fly pupae

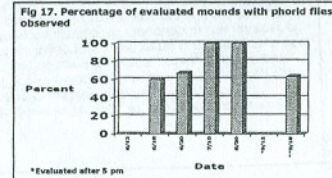


Fig 17. Percentage of evaluated mounds with phorid flies observed

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Thanks to Bill Kinkaid, UA-CES Pine Bluff for GPS assistance; the USDA-ARS Lab for the materials used in releases; David OI, USDA-ARS Gainesville, FL for on-site support; Sanford Porter, USDA-ARS Gainesville, FL for supplying phorid flies and consultation; and USDA PPQ for supplying materials and phorid flies for the Bradley Co. release





# Morphological Embryonic Development of the Red Imported Fire Ant *Solenopsis invicta* Buren (Hymenoptera: Formicidae)

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## Abstract

Embryonic characteristics during embryonic development was studied by external observation of the red imported fire ant *Solenopsis invicta*. Early embryonic patterning belonged to the long-germ-type. The germ band formed at the ventral side of the egg, composed of the incipient head lobes, the gnathal and thoracic region, and a growth zone. The germ band developed by extending and curving dorsally in both anterior and posterior directions, until the head and the tail almost connected with each other on the dorsal side of the egg when the germ band reached its maximum length. In the mean time, the segmentation and appendages appeared on the germ band. Five distinctive embryonic stages were evident according to the external characteristics of the growing embryos.

## Introduction

Hymenoptera is one of the best investigated insect orders in the field of embryology. Most of the subjects studied are in the families: Tenthredinidae, Trichogrammatidae, Ichneumonidae, Braconidae, and Apidae. However, little information is available on the embryology of the Formicidae.

The red imported fire ant, *Solenopsis invicta* has become a major pest since its introduction into the United States. Its damage and substantial impact on people, animal, and agriculture are growing because of its aggressive nature. Though intensive studies have been done on its biology, no research has been conducted on its embryology which may help to expand our knowledge of its community structure and management during the early stage of its life cycle.

The present paper reports the external characteristics of *S. invicta* embryos during the process of embryonic development.

## Materials and Methods

Emerged *S. invicta* alates were collected on the campus of Auburn University in May 2002. Alates were kept in petri dishes containing moist paper towel and food source, and maintained in an incubator at 25 °C, 95% RH, in total darkness. Eggs were collected daily two days after alates collection.

The eggs were fixed in Carnoy's solution for about 3–5 minutes before being transferred to 90% alcohol. Egg development was observed and photographed under a stereo-microscope.

Fifteen newly laid eggs were measured for length and diameter at the widest point, using a micrometer and a stereo microscope.

## Results

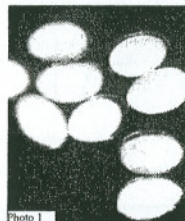


Photo 1. Newly laid eggs are ellipsoid, with a slightly rounded anterior end and pointed posterior end,  $0.45 \pm 0.02$  mm in length and  $0.28 \pm 0.02$  mm in maximal width.

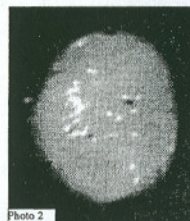


Photo 2. Developmental stage 1: Long germ band forms and develops at the posteroventral side of the egg.



Photo 3 and 4. Developmental stage 2: Germ band extends in both anterior and posterior directions: the posterior end curves around the posterior pole of the yolk mass onto the dorsal surface, and the anterior end immerses anterodorsally into the yolk mass. By this time, the segmentation is completed and the appendages appear.

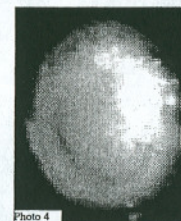


Photo 5 and 6. Developmental stage 3: Germ band begins to shorten and widen with the head and the tail on the dorsal surface moving towards the anterior and posterior pole of the egg respectively. The appendages on the head grows longer, but those on the thorax and abdomen stay unchanged.

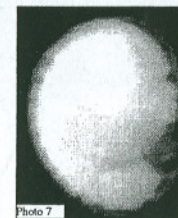


Photo 7. Developmental stage 4: The head of the embryo rotates from the dorsal side to ventral side of the egg and the dorsal closure of the embryo continues.



Photo 8 and 9. Developmental stage 5: The embryo begins to mature after dorsal closure and takes the form of a larva with the head directed backwards. At this time, the larva is ready to hatch from the anterior end of the egg.



## Discussion

- The early embryonic patterning of *S. invicta* was of the long-germ-type, and the embryonic primordium was long and composed of the incipient regions that would produce the head lobes, gnathal parts, thorax, and the abdomen.
- The blastokenesis of *S. invicta* was more distinct than that of other Hymenopteran insects previously investigated. It consisted of three successive embryonic movements: elongation of the germ band, shortening and widening of the germ band, and the displacement of the head anteriorly.
- In Hymenoptera, the extension of the germ band was generally exhibited at the posterior end, but in *S. invicta*, it was proceeded in both anterior and posterior directions. The posterior end of the germ band curved around the posterior pole of the yolk mass onto the dorsal surface, and the anterior end immersed anterodorsally into the yolk mass. The latter is not found in the embryogenesis of other Hymenopterans.
- In other insects, during the process of dorsal closure, it is usual for the embryo to take in all the yolk materials. However, it was interesting that there were some yolk balls left in the outside of the head region, when most of the yolk materials were enclosed within the embryo.



# Release and Establishment of the Fire Ant Decapitating Fly, *Pseudacteon tricusps*, in the Southeastern United States

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Between 1996 and 2002 the decapitating fly, *Pseudacteon tricusps*, was released at 56 sites in the southeastern United States (Fig. 1) as a self-sustaining biocontrol agent of the red imported fire ant, *Solenopsis invicta*. Most releases have been made by the USDA-ARS, CMAVE, Gainesville, FL (32 sites) and the University of Texas, Austin (15 sites). In the spring of 2002, USDA-APHIS funded a joint rearing effort (USDA-ARS, USDA-ARS, and the Florida Department of Agriculture) that has taken over rearing and release responsibilities from USDA-ARS. Overwintering populations of flies were successfully established at 19 sites in 6 states (AL-3, FL-5, LA-3, MS-1, SC-2, TX-5). Fly populations at 10 of these sites have expanded a mile or more away from their release sites (Table). Evaluation of recent releases are still in progress at an additional 19 sites and releases appear to have failed at 16 sites. Fly populations from releases around Gainesville, FL fused in 2001 and were expanding outward at the rate of 10-20 miles a year in the fall of 2001 (Fig. 2). We currently estimate that they are coast to coast in North Florida and beginning to move into Georgia. Fly populations near Auburn, AL and Bonita Springs, FL also have begun to expand explosively. Studies of fly impacts are currently underway in FL, LA, AL, MS, TX, OK, and SC. We are rearing two additional species of decapitating flies (*Pseudacteon curvatus* and *Pseudacteon littoralis*) that should be ready for trial field releases on red imported fire ants in the spring of 2003.

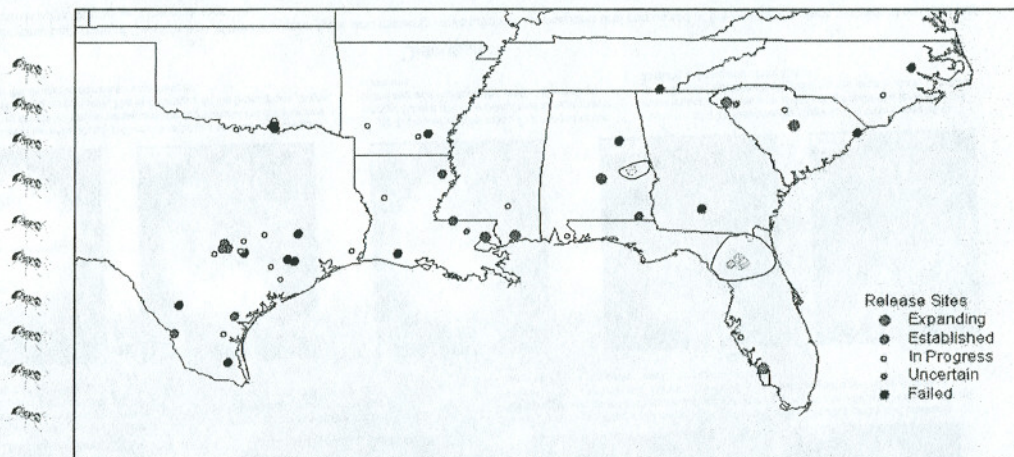


Fig. 1. Releases of the Brazilian decapitating fly, *Pseudacteon tricusps* in the southeastern United States (1996-2002).

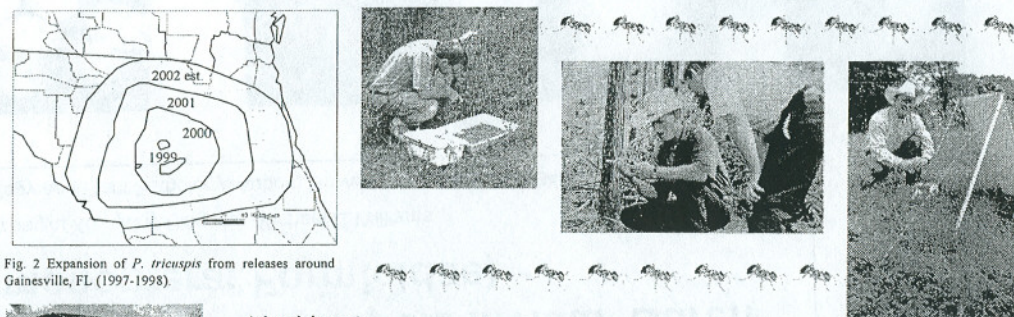


Fig. 2 Expansion of *P. tricusps* from releases around Gainesville, FL (1997-1998).

## Acknowledgements

The releases reported in this poster could not have been done without the help of numerous people in each of the states. Everyone that deserves to be acknowledged cannot be mentioned, but the following people were key to releases in their respective states: Kathy Flanders, Henry Dorough (AL), Lynne Thompson, Donna Shanklin, Kelly Loftin (AR), Lloyd Davis, Cynthia Vann (FL), Wayne Gardner, Stan Diffie (GA), Don Henne (LA), Kathleen Kidd (NC), Wayne Smith, J.T. Vogt (OK), Clyde Gorsuch, Tim Davis, Janet Kintz (SC), Roberto Pereira, Karen Vail (TN), Charles Barr, Jerry Cook (TX).



# Potential Global Range Expansion of the Red Imported Fire Ant, *Solenopsis invicta*

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**Abstract:** The red imported fire ant, *Solenopsis invicta* Buren, is an invasive pest that has become widespread in the southern United States and Caribbean after introduction from South America in the 1930's. This species has diverse detrimental impacts on recipient communities. It was recently discovered in Australia and New Zealand and has the potential to colonize numerous other regions. We used a dynamic, ecophysiological model of colony growth (Korzukhin et al. 2001) to predict the potential global range expansion of this invasive species. Based on minimum and maximum daily temperatures, the model estimates colony alate production and predicts future geographic range limits. Because *S. invicta* populations are limited by arid conditions as well as cold temperatures, we superimposed precipitation data upon temperature-based predictions, to identify regions that do not receive enough rainfall to support this species across the landscape. Many areas around the globe, including large portions of Europe, Asia, Africa, Australia, and numerous island nations, are at risk for *S. invicta* infestation. Quarantine officials

should be vigilant for any accidental introductions of this pest in susceptible regions. Costs of eradication increase dramatically as the area of infestation grows, and large infestations may be impossible to eradicate. Other South American *Solenopsis* fire ants (e.g., *S. richteri* Forel) may become invasive if the opportunity arises, and our predictions for *S. invicta* may approximate the potential range limits for these species as well. Manuscript in review in *Biological Invasions*. More detailed maps can be viewed online at [http://cmave.usda.gov/ifa/ifa\\_home.html](http://cmave.usda.gov/ifa/ifa_home.html)

Korzukhin MD, Porter SD, Thompson LC and Wiley S (2001) Modeling Temperature-Dependent Range Limits for the Fire Ant *Solenopsis invicta* (Hymenoptera: Formicidae) in the United States. *Environmental Entomology* 30: 645-655



**Figure Legend.** Symbols correspond to locations of weather stations (n=3,421) providing at least 5 years of reliable data. Red circles indicate areas of certain reproductive success; yellow triangles indicate areas of possible reproductive success; white circles indicate areas of unlikely reproductive success, based on temperature. Green shading indicates regions with sufficient annual precipitation (estimated at >510 mm) to sustain *S. invicta* across the landscape; olive green shading indicates arid regions that likely have insufficient rain (~510 mm annual precipitation). *Solenopsis invicta* will survive, however, in arid regions that are irrigated or near natural water sources.



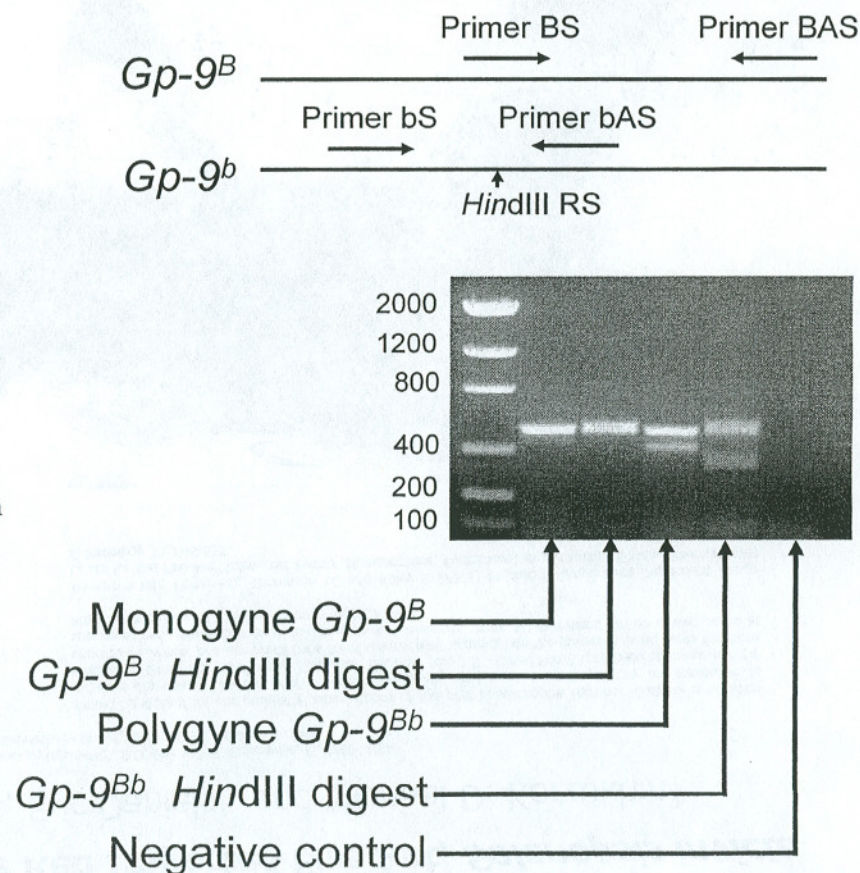
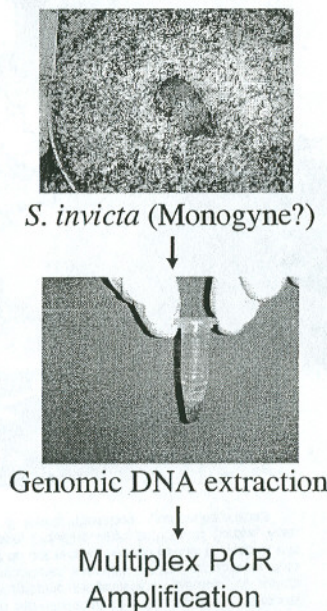
# IDENTIFICATION OF POLYGYNE AND MONOGYNE FIRE ANT COLONIES (*SOLENOPSIS INVICTA*) BY MULTIPLEX PCR OF GP-9 ALLELES

Steven M. Valles and Sanford D. Porter

USDA-ARS, Center for Medical, Agricultural and Veterinary Entomology

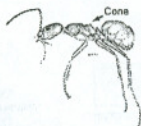
## Summary

Oligonucleotide primers were designed to discriminate between the *Gp-9B* and *Gp-9b* alleles found in monogyne and polygyne colonies of fire ant, *Solenopsis invicta*. Primers specific for the *Gp-9B* allele produced a 517 bp amplicon and primers specific for *Gp-9b* allele produced a 423 bp amplicon. When both sets of primers were multiplexed, homozygous monogyne ants produced a single 517 bp amplicon (specific for *Gp-9B*), whereas heterozygous polygyne ants produced one 517 bp amplicon and one 423 bp amplicon (specific for *Gp-9B* and *Gp-9b*, respectively) which allowed the *Gp-9* alleles to be discerned in a single reaction. This method was tested on ants from 20 monogyne colonies and 20 polygyne colonies and was 100% accurate in discriminating the two forms.





# PRELIMINARY ASSESSMENT OF *DORYMYRMEX* AND *SOLENOPSIS* INTERACTIONS



• Calixto, Marvin K. Harris, Allen Knutson and Charles Barr.  
Department of Entomology, Texas A&M University College Station, TX 77843  
acalixto@tamu.edu

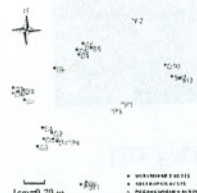


## INTRODUCTION

Hung (1974) made preliminary observations on fire ants (*Solenopsis invicta*) recovered from a single refuse pile of the Pyramid ant *Conomyrma* (= *Dorymyrmex*) *insana* and found fire ants predominated (43%); some authors (Smith 1965; Nickerson et al. 1975; Wilson et al.) suggested *Dorymyrmex* may kill newly mated fire ant queens, alate males and workers, and that perhaps it sometimes nests at the edge of the fire ant mounds, but these observations have not been quantified. The role of native ants in habitats treated for fire ants is not well understood. More knowledge of the interactions of fire ants and native ants is needed. We observed fire ant remains adjacent to *D. flavus* nests in a commercial pecan orchard in Burleson Co., TX while conducting extensive investigations on fire ant in pecan. The *D. flavus* nests were located in plots previously treated with Extinguish® bait (0.5% methoprene applied at 1.5 lbs/acre, on May 19, and October 12, 2000 and June 12 in 2001) and had fire ant nests nearby. Fire ant nest density had been reduced in these plots by about 70%. We documented the observations by collecting the midden (a raised, loosely aggregated mound of earth, sticks, dead ants, ant parts, and other materials constructed at the nest entrance by *Dorymyrmex*) from each of ten nests on July 31, 2002 and again on Aug. 8, 2002. Middens were inspected with the aid of a binocular microscope (4-60x) searching for arthropod remains and other materials that might be of interest. More than 98% of the arthropod remains found consisted of fire ants, with the initial inspection on July 31 showing an average minimum of 203 dead fire ants per midden and the Aug. 8 inspection showing 40 per midden. Further investigations are being conducted to determine how the fire ants came to be in the middens and whether or not these findings are important in fire ant management.

## MATERIALS AND METHODS I

- Study site: Commercial pecan orchard at Mumford, TX (Robertson Co.). (30° 44' 55" N; 96° 33' 12" W).
- 10 middens of *Dorymyrmex flavus* were collected randomly in an area previously treated for fire ants with methoprene bait Extinguish® (IGR) (applied on May 19, and October 12, 2000 and June 12 in 2001).



GPS PLOT OF ANT NESTS

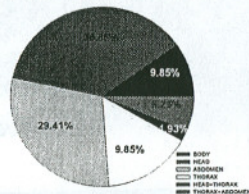
- The entire midden was collected from each nest on July 31, 2002 and nests were marked and referenced with GPS; middens were again collected on August 8 2002 from the same nests.

## MATERIALS AND METHODS II

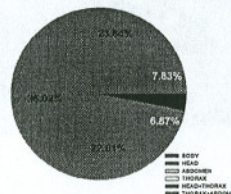
- The middens were placed in plastic snap cap vials (7 dram) and returned to the lab. Each midden was initially divided into 1.5 ml subsamples by volume. Then each subsample was inspected, cleaned of debris and soil particles, and all insect remains were sorted and preserved in alcohol. Arthropods and body parts were placed in vials with 70% alcohol. Ants were identified and counted, with body parts classified as:
  - Body= if body is complete; head-thorax-abdomen
  - Head= if only head is visible
  - Thorax= if only thorax is visible
  - Abdomen= if only abdomen is visible
  - Head + Thorax= if only head and thorax is visible
  - Thorax + Abdomen= if only thorax and abdomen is visible
- Parts of previously identified fire ants and native ants were used to compare and identify remains.
- Time spent on each sample on cleaning, sorting, etc. = 40 minutes
- Time spend on counting remains in each subsample: 45 minutes

## RESULTS AND DISCUSSION I

SOLENOPSIS INVICTA IN TEN DORYMYRMEX MIDDENS  
TOTAL BODIES = PARTS= 4870 (JULY 31 2002)

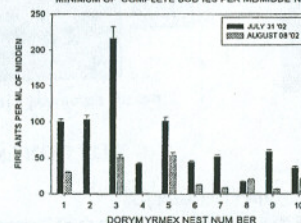


SOLENOPSIS INVICTA IN TEN DORYMYRMEX MIDDENS  
TOTAL BODIES = PARTS= 2148 (AUGUST 8 2002)



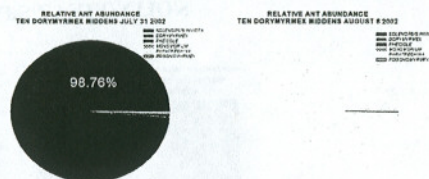
Fire ant segments (separate head, thorax or abdomen) predominated in *Dorymyrmex* middens on both sample days. Whether *Dorymyrmex* ants preyed upon the fire ants or just collected them remains unclear.

SOLENOPSIS INVICTA IN TEN DORYMYRMEX NESTS  
MINIMUM OF COMPLETE BODIES PER MUMMIDE N



Fire ants collected and recovered from each nest (July 31 '02: AVG= 77.24, SE:18.15; August 8 '02: AVG=20.54; SE:6.11)

## RESULTS AND DISCUSSION II



Body remains of five ant species were found on middens; *Solenopsis invicta*, *Dorymyrmex flavus*, *Pheidole* sp., *Monomorium minimum*, *Paratrechina* sp. and *Pogonomyrmex barbatus*. Note fire ants predominated.

## CONCLUSIONS

- *Solenopsis invicta* is the most common species occurring in *Dorymyrmex* middens. Body parts (head, thorax or abdomen) were more abundant than entire ants, but handling may have fragmented intact ants before inspection.
- The differences between sample days suggest that *Dorymyrmex* ants accumulate fire ants at a high rate over a short time period. If they are preying upon fire ants, this may indicate their potential in biological control.
- Other native ant species including *Dorymyrmex* do not occur in significant numbers in the middens.
- Further studies are necessary to understand relationships among ant species and evaluating their potential as biological control agents. When combined with bait applications, *Dorymyrmex* seems to tolerate bait treatments (being little affected by the IGR) and second by competing with fire ants.

## References

- Hung, A. C. F. 1974. Ants recovered from refuse pile of the Pyramid ant *Conomyrma* *insana* (Buckley) (Hymenoptera: Formicidae). Ann. Entomol. Soc. Amer. 67(3):522-523.
- Nickerson, J. C., W. H. Whitcomb, A. B. Bhatkar and M. A. Navas. 1975. Predation of founding queens of *Solenopsis invicta* by workers of *Conomyrma* *insana*. Florida Entomol. 58:75-72.
- Smith, M. R. 1965. House infesting ants of the eastern United States. U.S.D.A. Tech. Bull. 1326. 105p.
- Weisen, N. L., J. H. Diller and G. P. Markin. 1971. Foraging territories of imported fire ants. Ann. Entomol. Soc. Amer. 64:600-65.

## Acknowledgments

- Dash Bhagirathi provided lab help.
- Partial Funding was provided by the TEXAS FIRE ANT INITIATIVE and USDA Special Grant

More information about FIRE ANTS

• <http://fireant.tamu.edu>





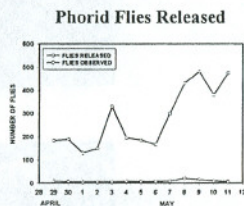
# AREAWIDE SUPPRESSION OF FIRE ANTS - TEXAS HIGHLIGHTS OF YEAR 2002



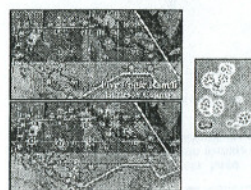
Charles Barr, Alejandro Calixto and Bart Drees  
Texas Cooperative Extension - Texas Agricultural Experiment Station  
Department of Entomology, Texas A&M University System, College Station, TX 77843-2475  
c-barr@tamu.edu



## Five Eagle Ranch - Burleson Co. BIOLOGICAL CONTROL SITE



### Natural *Thelohania* spread



## MOUND COUNTS (Treatment applied on May 31)



1,475 mounds; AVG=21.6; STD: 14.73. **Treated** (0 photo): 538 mounds; AVG=26.76; STD: 13.13. **Control** (0 photo): 940 mounds; AVG=31.33; STD: 15.43.



421 mounds; AVG=9.23; STD: 8.30. **Treated** (0 photo): 44 mounds; AVG=1.2; STD: 4.25. **Control** (0 photo): 397 mounds; AVG=13.23; STD: 7.7.



630 mounds; AVG=12.4; STD: 11.47. **Treated** (0 photo): 42 mounds; AVG=1.1; STD: 2.40. **Control** (0 photo): 588 mounds; AVG=19.6; STD: 9.57.

## FORAGING EVALUATION



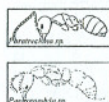
2,079 Fire Ants apprx.; AVG=40.50%; STD: 2.48. **Treated** (0 photo): 1203 IFA foraging; AVG=42.63%; STD: 13.49. **Control** (0 photo): 1426 IFA; AVG=47.54%; STD: 24.33.



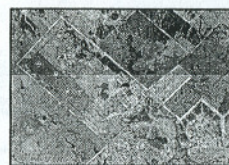
1794 Fire Ants apprx.; AVG=14.89%; STD: 24.18. **Treated** (0 photo): 105 IFA foraging; AVG=2.54%; STD: 9.79. **Control** (0 photo): 1699 IFA; AVG=43.22%; STD: 12.44.

## PITFALL TRAPS EVALUATION

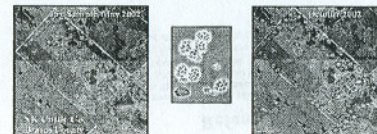
Total of 26,825 IFA captured on May; AVG=67.06; STD:108.44. 9 Species recorded being IFA the dominant (99.47%), followed by *Diplophoropium* (0.281%), *Paratrechina* (0.177%), *Dorymyrmex* (0.10%), *Tapinoma*, *Formica*, *Pachycondyla*, *Pogonomyrmex* and *Monomorium* sharing (0.003%).



## NK Ranch Co. - Brazos Co. CONTROL SITE



### Natural *Thelohania* spread



## MOUND COUNTS (Bait applied on October 11)



1264 mounds; AVG=24.08; STD: 19.14. **Treated** (0 photo): 198 mounds; AVG=13.75; STD: 6.94. **Control** (0 photo): 809 mounds; AVG=34.96; STD: 16.08.



484 mounds; AVG=8.88; STD: 7.7. **Treated** (0 photo): 40 mounds; AVG=0.2; STD: 3.09. **Control** (0 photo): 364 mounds; AVG=12.13; STD: 7.31.



807 mounds; AVG=16.14; STD: 16.20. **Treated** (0 photo): 180 mounds; AVG=4.65; STD: 19.45. **Control** (0 photo): 706 mounds; AVG=23.53; STD: 15.09.

## FORAGING EVALUATION



2188 Fire Ants apprx.; AVG=55.74%; STD: 13.24. **Treated** (0 photo): 1108 IFA foraging; AVG=55.30%; STD: 13.63. **Control** (0 photo): 1470 IFA; AVG=58.68%; STD: 12.95.



3099 Fire Ants apprx.; AVG=41.80%; STD: 21.20. **Treated** (0 photo): 232 IFA foraging; AVG=11.69%; STD: 19.86. **Control** (0 photo): 1858 IFA; AVG=61.30%; STD: 19.45.

## PITFALL TRAPS EVALUATION

Total of 12,618 IFA captured on May; AVG=31.54; STD:49.74. 5 Species recorded being IFA the dominant (99.76%), followed by *Monomorium* (0.012%), *Paratrechina* (0.09%), *Pogonomyrmex* (0.007%) and *Hypoponera* (0.007%).





# The use of community participation in surveillance for RIFA in South East Queensland, Australia



Michelle Milzewski, Manager Community Engagement  
Jenny Bibb, Manager Public Relations  
Fire Ant Control Centre Brisbane – Australia

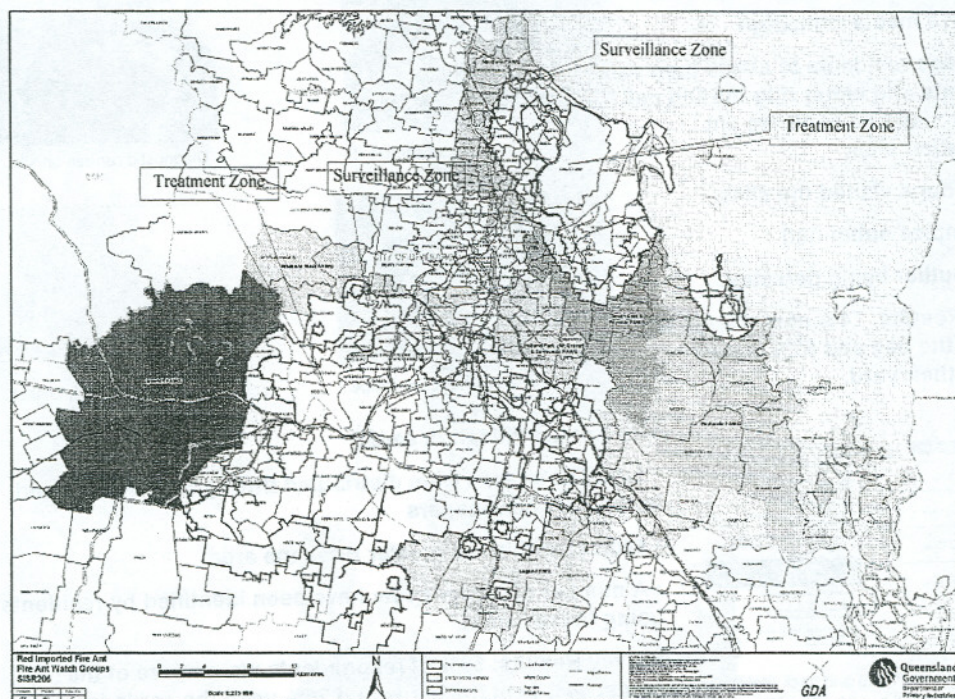


**Challenge:** To find any undetected fire ant nests outside the existing Treatment and Surveillance Zones.

**Fire Ant Community Watch Group Initiative** – a partnership between government and community.

Seven groups are established across the greater Brisbane area with more to follow. Groups are comprised of individuals and representatives of other community and environmental groups. FACC provides training, secretarial and coordination support. Groups conduct surveillance in parkland areas and assist in maintaining community support through public education.

Map showing areas covered by each Fire Ant community watch group in relation to the treatment and surveillance zones of the fire ant eradication program.



Over 50 parkland areas have been checked for fire ants.



Fire ant watch groups maintain community awareness and support.



There are over 180 Fire Ant Rangers.



# The use of public relations activities in passive surveillance for RIFA in South East Queensland, Australia



Jenny Bibo, Manager Public Relations  
Michelle Milzewski, Manager Community Engagement  
Fire Ant Control Centre Brisbane – Australia



**Challenge:** To find any undetected fire ant nests outside the existing Treatment and Surveillance Zones.

Passive surveillance promoted and encouraged through public relations activities including distribution of Identification cards and Find the Fire Ant Day.



**Find the Fire Ant day – 28 July 2002**

- extensive media campaign
- encourage residents to check their yard for fire ants and either ring the DPI call centre or visit one of 10 fire ant identification sites
- 2609 groups visited the sites,
- 341 samples submitted
- 1 new outlier being detected.

**Survey Results:** 74% of residents were aware of the day and 52% actually checked their yard.



Information and sample ID site.



Diagnostic centres on site.



Extensive media coverage.



Map showing 10 info sites.



Fire Ant ID card front (left) and back (right). Actual size 10.5 x 7.5 mm (4 x 3 inches).

## Fire Ant identification cards

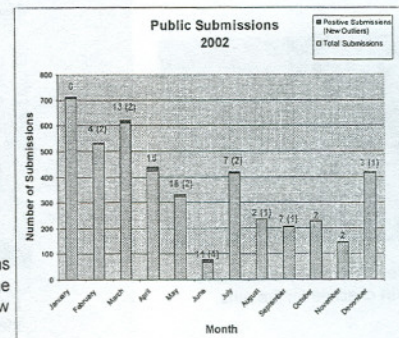
- 1.4 million cards were distributed throughout Queensland in regional newspapers
- 540,995 within the greater Brisbane area.
- A number of outlier nests have been identified by residents using these cards.

**Survey Results:** 59% of respondents were aware of the cards, 44% kept the cards and 26% used the cards to check their yards.

## Passive Surveillance program results:

- 27 outliers detected outside the treatment zone
- 15 outliers detected by public
- 7638 sample submissions by public
- 72,500 calls to call centre
- 85,500 visits to website

Fire Ant sample submissions by the public in 2002 and the number of positives and new outliers found.





# Diagnostics & its role in the fire ant eradication program in Brisbane - Australia.



Marlene Elson-Harris, Kym Johnson, Shane Moloney & Lynne Griffin  
Fire Ant Control Centre Brisbane – Australia



Diagnostics underpins all operational units within the Fire Ant Control Centre with accurate and timely identifications.



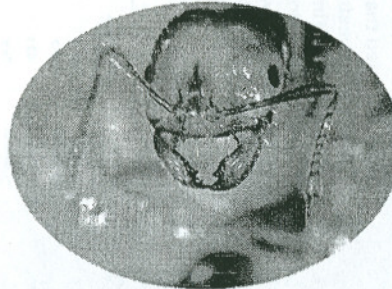
On-site identifications at various centres throughout the greater Brisbane Area.

Entomological training for community watch groups.



Spin-offs from Diagnostics:

Early detections & eradication of otherwise undetected incursions including Argentine ants *Linepithema humile*, Tropical fire ants *Solenopsis geminata* (above left) and Crazy ants *Anoplolepis gracilipes* (above right).



All ant samples generated by the program are identified by the diagnostics unit.

Since February 2001 > 31400 samples have been identified.

To date 9% of these have contained *Solenopsis invicta*.

Up to 1200 samples per week have been processed by this unit.

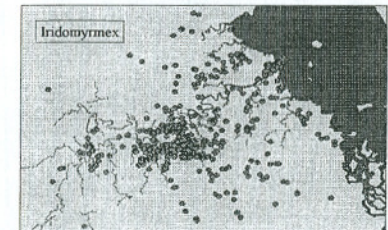


*Solenopsis invicta*



Providing scientific support for public relation events.

Ensuring accurate scientific information for community engagement meetings.



This comprehensive survey has enhanced our knowledge of native ant distributions in south east Queensland. Currently 52 of 103 known Australian genera have been recorded.



# Increasing the treatment options for fire ants in Brisbane - Australia



John R. Hargreaves & John Johnston  
Fire Ant Control Centre Brisbane – Australia



Specific niches have occurred within the treatment programme that have allowed us to expand our treatment options.

## Hay bales

Although methyl bromide is registered for use against ants at 120g/bale for mulching straw or hay, there was no registration for hay in storage for fodder. However, the general fumigation rate of 32g/cubic meter gave excellent control of RIFA. The National Registration Authority (NRA) has ratified this use as permit No 5168.

Sequence	% Mortality	% Efficacy at 95% Confidence Level
Bale insertion without fumigation	2.3	
Fumigation 9 – 10 Oct 01	100	99.84
Fumigation 11-12 Oct 01	100	99.69
Fumigation 16 – 17 Oct 01	100	99.78
Total Fumigated	100	99.93

Sampling interval Pasture	Site 1	Site 2	Site 3	Site 4	Mean and Standard error
2 hrs after	0.364	0.103	0.302	0.047	0.204±0.066
1 day	1.332	0.910	0.000*	0.172	0.603±0.271
3 days	0.581	0.379	0.922	0.079	0.490±0.153
7 days	0.318	1.365	0.000*	0.104	0.420±0.280
14 days	0.028	0.095	0.042	0.000*	0.041±0.017
28 days	0.000* **	0.000* **	0.000*	0.000*	0.000*
Soil					
2 hrs after	3.148	0.484	0.079	0.087	0.949±0.640
1 day	0.670	0.746	0.093	0.504	0.503±0.126
3 days	0.712	0.344	0.000*	0.051	0.277±0.095
7 days	1.196	0.123	0.008	0.000*	0.332±0.251
14 days	0.103	0.019	0.000*	0.009	0.033±0.021
28 days	0.010 **	0.000* **	0.000*	0.000*	0.000*

S-methoprene residues expressed in mg/kg dry weight from grazing locations within the treatment area for fire ant baiting.

## S-methoprene residues on pastures

The study provided valid S-methoprene residue data to the National Registration Authority (NRA) for the treatment of pasture. A suitable withholding period for grazing beef cattle could consequently be established. It was calculated that 1mg S-methoprene / kg dry weight of pasture as 100% of a beast's diet was the maximum feeding rate. The data from the trials were submitted to NRA. A permit, (No.5998) was issued allowing a nil withholding period for grazing animals.

## Protective treatment for nursery stock

A slow release formulation of chlorpyrifos is used in the nursery trade as a control for beetle larvae (black vine weevil) at the usage rate of 1kg/m<sup>3</sup> potting mix. We know from previous trials that RIFA workers can be controlled at 10-12mg chlorpyrifos / kg potting mix. The table (right) shows the decay in a production glasshouse over 6 months. An application for an interim permit for 6 months protection has been forwarded to NRA. Data to date still shows levels of 42 – 51mg / kg in mix exposed for 12 months.

Date Tested	Chlorpyrifos mg/kg	% Mortality suSCon	% Mortality Untreated
30Jan02	61.0	100	6.7±0.4
20Feb02	87.2	100	4.7±2.2
20Mar02	102.1	100	4.2±2.2
19Apr02	78.9	100	4.1±1.7
22May02	82.6	100	1.1±1.5
21Jun02	83.6	100	2.1±1.1
17Jul02	51.3	100	7.0±0.8
22Aug02	109.8	100	5.9±1.03

Residues and mortality of RIFA workers exposed to the collected mixture.

Treatment	Conc	Pretreat	3DAT	7DAT	14DAT	21DAT
nil		2.8a	2.5a	2.7a	2.7a	2.8a
fipronil	1L / nest	2.8a	2.8a	2.6a	2.5a	2.6a
fipronil	2L / nest	2.8a	2.6a	2.0a	1.4b	1.4b
fipronil	4L / nest	2.7a	1.5b	0.8b	0.4c	0.1c

Treatment	Conc	Pretreat	1DAT	2DAT	3DAT	7DAT	14DAT
nil		3.0a	3.0a	3.0a	3.0a	2.9a	2.9a
chlorpyrifos	20ml / 100L	2.9a	0.1b	0.0b	0.0b	0.0b	0.0b
fipronil	2.5ml/100L	2.9a	2.4c	1.4c	1.0c	0.7c	0.1b

## Fipronil

Trials of fipronil as a drench have shown it to give similar levels of control to the chlorpyrifos standards. Reducing the volume of wash of fipronil from 4L / nest has been significantly less effective. Application for use as a nest injection and drench has been made to NRA.

Mean activity rating per nest after 4L wash had been poured on the nest and in a 1m diameter circle around the nest. Work conducted at Swanbank Qld, October – November, 2002.

## Compost treatment of infested soil

Recordings of temperatures in composting heaps of commercial facilities have shown that some reach a consistently high temperature (over 65 degrees C), very rapidly. With good storage facilities, this treatment is being considered as a RIFA disinfestation procedure.





# Progress of the Fire Ant Eradication Program in Brisbane - Australia



Craig Jennings, Technical Services  
Fire Ant Control Centre Brisbane – Australia

## Treatment

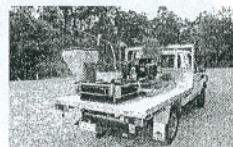
Our aim is to bait the treatment area (green zone on map) 12 times over three years

We are up to the 7<sup>th</sup> round

The treatment area is 41 957 hectares  
102678 land parcels

This has involved 89 586 consents from  
residents to enter their properties

Only 605 residents have refused treatment –  
these undergo continual surveillance



Aerial application: 53%  
All Terrain Vehicles application: 22%  
Manual/foot application: 25%

180 259 hectares have been treated since the beginning of  
the program - this includes 363 963 land parcels

75% of properties had no active nests during a survey of  
900 known infested premises

Monitoring sites show a 90% overall reduction in nest  
density

Large areas of the treatment zone have been free of RIFA  
since February 2002



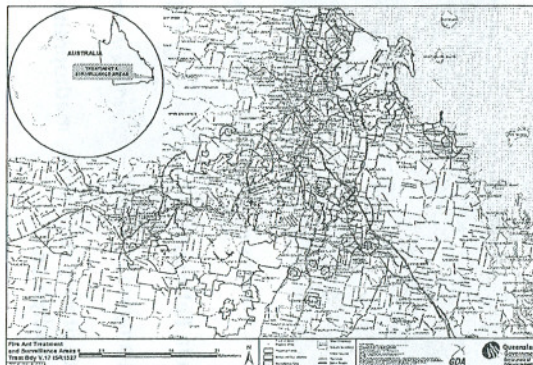
## ERADICATION

The aim of the Fire Ant Control Centre is to eradicate  
Red Imported Fire Ants

They were first discovered in Brisbane Feb 2001

Program commenced 24 September 2001

The eradication program involves a multidisciplinary  
approach that includes treatment, surveillance, PR,  
community engagement, risk management and security  
and science



### FACC Eradication Plan Schedule

	Year					
	2001	2002	2003	2004	2005	2006
Surveillance						
Passive						
Active						
Post treatment						
Treatment						
Scheduled						
Nuisance						

Community based surveillance

Outside the Treatment Zone

Within the Treatment Zone

3-4 Treatments per year

For persistent colonies

## Surveillance

Our aim is survey all land in the surveillance zone (Yellow zone on  
map) at least once a year

Enhanced surveillance zones (Orange zone on the map) around  
outlying infestation will be done twice a year

Outside of the surveillance zone we will be undertaking targeted  
surveillance (based on probability of infestation)

Targeted surveillance will include areas such as new housing  
developments



44 289 hectares have been surveyed outside the treatment  
zone

This area consists of 136 308 land parcels including 112 165  
properties

73 infested properties in 27 outliers have been detected beyond  
the treatment zone

In the 100% surveillance zone the occurrence of infested  
properties is 0.031% or <1 in 3000

60 of these infested properties have fewer than 5 nests

10 outliers are beyond the 100% surveillance zone

We believe that we are close to delineating the extent of the  
RIFA infestation in Queensland





# Ecological effects of *Solenopsis invicta* in Brisbane - Australia



Tania Fuessal†, Adriana Najar†, Jo-anne Holley\* and Kris Plowman\*  
Fire Ant Control Centre Brisbane – Australia



## Assessment of ecological effects of *Solenopsis invicta* and the treatment program on soil and litter invertebrates

Soil invertebrate and mobile ground invertebrate assemblages were sampled in 2002 in sites:

- within the treatment zone with *S. invicta* - TFA
- within the treatment zone and not colonised by *S. invicta* - TNFA
- outside the treatment zone - Control

Treatment began in September/October 2001.



Examples of the sites sampled are shown above.

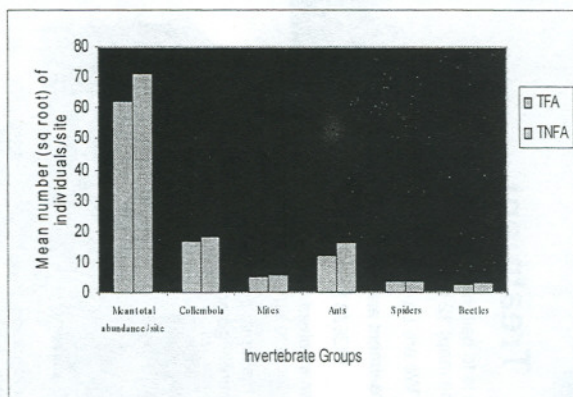
Soil cores (5 x 5 cm) taken in February and extracted by Tullgren funnels provided samples of soil invertebrates.

Mobile ground fauna was sampled by pitfall traps (10 x 7 cm) between May and July.

Treatments included: insect growth regulators, methoprene & pyriproxyfen; and the metabolic inhibitor, hydramethylnon.

Preliminary results suggest that the abundance of mobile ground dwelling invertebrates tends to be greater in treated areas without fire ants than in treated areas with fire ants.

The abundance, diversity and functional groups of beetles and ants recorded in the pitfall traps, and the abundance and diversity of soil dwelling Oribatid mites, is being estimated. These estimates are used to assess the impact of fire ant infestations and the treatment regimes on invertebrate assemblages.



† Tania Fuessal (Griffith University); Adriana Najar (University of Queensland); \* Ecology, Scientific Services, Fire Ant Control Centre.



# Monitoring the fire ant eradication program in Brisbane - Australia



Evan Harris, Stuart Mutzig & Paul Garland  
Fire Ant Control Centre Brisbane – Australia

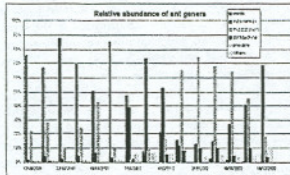
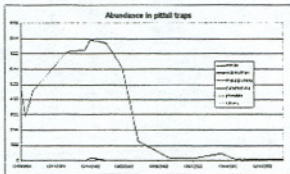


Post treatment monitoring and assessment of treatment regimes throughout infested areas using population monitoring techniques and brood/nest assessments.

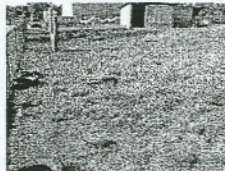
Population monitoring to assess the numbers, nest density and rate of decline in active infestations. Currently 66 sites are regularly assessed.



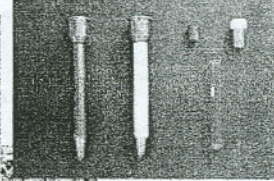
Current distribution of monitoring sites.



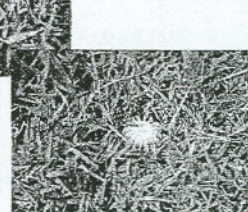
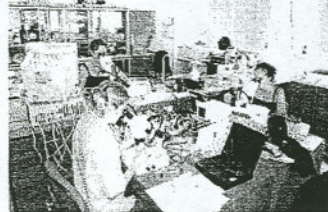
Typical data analysis of a monitoring site.



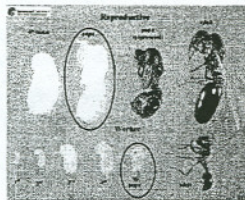
Semi-rural monitoring site.



Pitfall trap components.

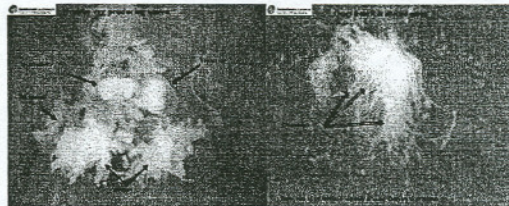


**Brood and nest assessment:** This technique is used to determine the reproductive status of active nests which reveals the effects of the treatment on these areas where population data and surveillance techniques cannot. Presence of worker ants may not necessarily indicate a healthy nest capable of producing viable alates and continuing the infestation.



Active nests are opened and the brood searched for presence of worker pupae. Queens and dealates are harvested for dissection and the presence of other invertebrates in the nest are recorded.

Dissection indicates the insemination and ovary status of those harvested allowing a rating of their viability to be made.



An area of major infestation in suburban Brisbane showing locations of positive RIFA.



The same area showing extent of infestations revealed by property survey.



The area again overlaid with brood assessment results.



# Pastoral Peace?

## A third year report on *Thelohania solenopsae* in a Mississippi coastal pasture.

S. James<sup>1</sup>, A.M. Callcott<sup>1</sup>, H. Collins<sup>1</sup>, D. Oi<sup>2</sup>, R. Weeks<sup>1</sup>, and D. Williams<sup>2</sup>

<sup>1</sup>USDA, APHIS, PPQ, CPHST, Soil Inhabiting Pest Laboratory, Gulfport, MS

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### INTRODUCTION

In many situations it is not economically feasible to chemically treat to achieve fire ant suppression. Infested agricultural lands, such as pastures often fall into this category and thus stand to reap great benefit from the introduction of biological controls for imported fire ants. The microsporidium *Thelohania solenopsae* (Microsporidia: Thelohaniidae), documented infecting red imported fire ants (*Solenopsis invicta*) in Brazil (Knehl et al. 1977) and black imported fire ants (*S. richteri*) in Argentina, has been determined by USDA, ARS, CMAVE to decrease colony populations and colony vigor (Brisson et al. 1993a, 1993b, 1994). Therefore, field trials have been initiated to determine the potential of *T. solenopsae* as a biological control agent here in the United States of America.

### OBJECTIVE

Our objective was to assess *Thelohania solenopsae* as a biological control agent for *Solenopsis invicta* in southern Mississippi.

### MATERIALS and METHODS

October 19, 1999 we assisted ARS with the initiation of a trial to evaluate field releases of the pathogen *Thelohania solenopsae*. Two sites in southern Mississippi, one polygyne in Hancock Co. and one monogyne in Harrison Co. were selected for the inoculation trials. The monogyne site, however, was lost early on due to pasture improvements. The four plots at each site were divided into two inoculation plots and two non-inoculated control plots. Due to the high density of mounds at the polygyne site, the circular test plots were only 1/4 acre in size instead of the standard 1/2 acre. Brood infected with *T. solenopsae* (field collected by ARS prior to study) was introduced in 3.5-g amounts to nine mounds within each of the inoculation plots. As weather and circumstance permit, plot evaluations consisting of number, population index, and within-plot location of mounds have been conducted every few months. Evaluations to date occurred at weeks 0 (pre-inoculation), 12, 20, 28, 36, 49, 76, 84, 92, 100, 109, 118, 127, 141, 155, 161, and 173. Repeated measures ANOVAs, with treatment, year, and season nested within year, were performed on both colony number and population index. Population indices with brood were scaled for a heavier weighting than those without. Due to site flooding from tropical storm Isadore and hurricane Lili in weeks 152 and 153, only data from weeks 0 - 141 are analyzed.

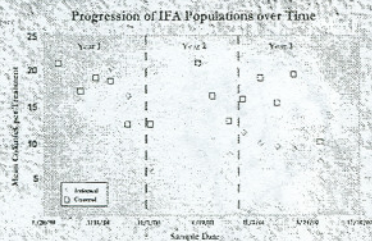
Worker samples were also collected from each mound during these evaluations and frozen until they could be examined. Ants from each sample were ground in a tissue grinder and wet mount slides were made of the resulting slurry. The slides were studied under 400x magnification on a phase contrast microscope for presence of *T. solenopsae* spores (Brisson et al. 1993a).

### RESULTS and DISCUSSION

#### Colony Mortality

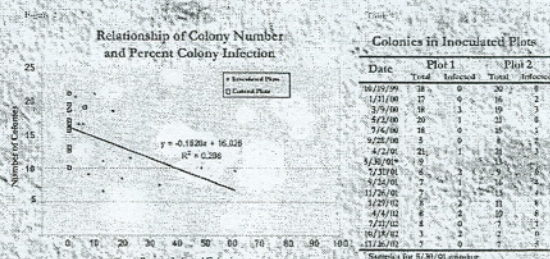
The numbers of colonies and population indices in plots were significantly influenced by treatment, year of the trial, and season within the year (df=55, F=4.481, P<0.001; df=55, F=5.956, P<0.0001 respectively). Colony mortality as measured in numbers of colonies and total population index per plot displayed much fluctuation within each year based on season (effects test, F=2.863; F=3.737) (Figure 1). While some colonies do succumb to summer heat, often ants travel deeper in the soil to avoid desiccation and thus are harder to adequately sample causing an apparent if not actual drop in populations. Peak populations, likewise, annually coincide with the optimal weather in spring sampling when new colonies arise and ants are most active.

Inoculated and control plots displayed similar colony numbers and population indices for the first year of the trial and to some extent in the second year. However, enough exposure to *T. solenopsae* infection occurred by the third year to cause inoculated plots to deviate from the seasonal cycling still demonstrated by the control plots (year effects test, F=9.224; F=10.441) (treatment effects test, F=8.843; F=14.235). Very few colonies were located in the area after the flooding that occurred a few weeks prior to the week 173 sampling. As of week 173 the number of control plot colonies had not returned to the level expected for that time of year. We will continue monitoring at least through 2003, but it is possible that the floods during fall of 2002 have disrupted the control plots at this site beyond further use.

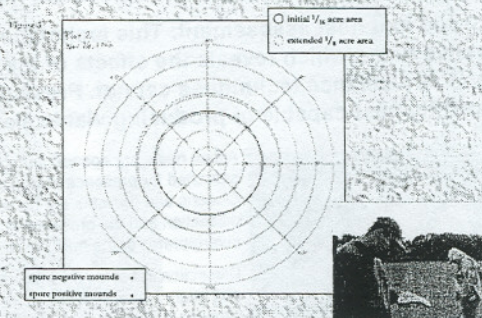


#### Presence of Pathogen

No spores were detected in pretreatment samples (table 1). At 12 weeks after inoculation, 2 mounds in one of the inoculated plots were positive for spores. By week 20, both of the inoculated plots had three positive colonies, and spores were detected from 2 mounds in one of the control plots. Since that week, no other spore positive results have been found for the control plots. The percent of infected colonies in both inoculated polygyne plots increased as the number of colonies decreased (figure 2). Highest numbers of spore positive colonies occurred in the third year while the inoculated plots were deviating from the seasonal population increase seen in the controls.



Among the properties of an ideal biological control agent is the ability to sustain itself in the field. Fluctuation of infection does appear in the data, as colonies recorded in the same location will test positive on one date, negative on another, and then positive again later on. Potential contributing factors include ability to detect non-spore forming stages of the disease, inadequate sample collection due to season, and colony movement in and out of the monitored plot. In an attempt to determine if flooding moved infected colonies out of the original plots, the week 161 sampling area was increased to 1/2 acre. One plot in this sampling had no positives within the original 1/4 acre plot but one positive colony at 0.3 in out of the original zone. The other inoculated plot doubled its number of positive colonies with five inside the original plot and five in the extended sample area (figure 3). Alterations in our sampling methods have been initiated in order to develop a more complete picture of *Thelohania* activity in the site.



Even in this limited sampling our results to date support the assessment of *Thelohania solenopsae* as a useful biological control. Within three years, significant reduction of imported fire ant populations and ability to sustain area infection have been demonstrated. The weather conditions and flooding of this site were not ideal for the maintenance of this experiment as it was initially designed, but they are typical of the coast and further monitoring, regardless of the continued status of the control plots, is certainly warranted.

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Direct questions or comments to: Shannon.S.James@aphis.usda.gov



abdomen is visible. Body remains of previously identified fire ants and native ants were used to compare and identify remains. The time used on cleaning up, sorting, and storing the samples was approximately 40 minutes and the time used on counting remains on each subsample was approximately 45 minutes.

## Results and Discussion

Fire ant segments (separate head, thorax or abdomen) predominated in *Dorymyrmex* refuse piles for both sample days. Whether *Dorymyrmex* ants preyed upon the fire ants or just collected them remains unclear. There were 6870 fire ant parts collected on July 31 and 2145 on August 8, they recovered 31% of fire ants parts in eight days being the heads the most frequent part found on the first sample day (28.47% ) and abdomens for the second sample day (7.40%) (Table 1). Pyramid ant nest where the complete midden was removed and no ant remains were left recovered a significant number of fire ants at a highly considerable rate during a short period of time (July 31 '02: AVG= 77.24; SE:18.15); August 8 '02: AVG=20.54; SE:6.11) (Figure 1). Body remains of five other ant species including pyramid ants were found on middens; *Dorymyrmex flavus*, *Pheidole* sp., *Monomorium minimum*, *Paratrechina* sp. and *Pogonomyrmex barbatus* but in extremely low numbers they don't represented more than 0.01% of the total samples, for both sample dates the highest recoveries were represented by fire ants in up to 99.9%. Somehow *Dorymyrmex* seems to be putting a lot of effort on "collecting" or "killing" fire ants and putting them on the refuse piles. If they are preying upon fire ants, this might be an indication of their role as potential biological control agent. Further studies are necessary to understand the interactions between *Dorymyrmex* and *S. invicta*, in particular case. when combined with bait applications. *Dorymyrmex* seems not to be affected by IGR bait treatments We speculate that *Dorymyrmex* in combination with IGR bait treatments may significantly extend the treatment interval required to maintain *S. invicta* densities below economic thresholds because *Dorymyrmex* interferes with *S. invicta* colonization and, even when established, interferes with *S. invicta* rate of increase to noxious densities, further and more detailed studies are needed.

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## Acknowledgments

Dash Bhagirathi provided lab help by sorting and cleaning samples. Partial Funding was provided by the TEXAS FIRE ANT INITIATIVE and USDA-ARS Areawide Suppression of Fire Ants Program.



	Body	Head	Abdomen	Thorax	Head+Thorax	Thorax+Abdomen
July 31	7.61±1.38	<b>28.47±8.01</b>	22.72±6.77	12.13±3.11	1.49±0.41	4.83±0.95
August 8	1.50±0.49	4.9±1.72	<b>7.40±2.12</b>	4.68±2.16	1.41±1.18	0.64±0.21

Table 1. Mean and average of fire ant remains recovered from *Dorymyrmex flavus* middens



## Pastoral Peace?

### A third year report on *Thelohania solenopsae* in a Mississippi coastal pasture.

S. James 1 , A.M. Callcott 1 , H. Collins 1 , D. Oi 2 , R. Weeks 1 , and D. Williams 2  
1 USDA, APHIS, PPQ, CPHST, Soil Inhabiting Pest Laboratory, Gulfport, MS 2 USDA-  
ARS/CMAVE, Gainesville, FL

## ABSTRACT

On October 19, 1999, *Thelohania solenopsae* (Microsporidia: Thelohaniidae) was introduced to nine polygyne red imported fire ant mounds in two plots located in a Mississippi coastal pasture. Ant samples were collected and mound number and population indices were recorded every few months for three years. Colony numbers and population indices for inoculated and control plots were not significantly different through the first two years even though second year values for inoculated plots were slightly lower than in the controls. Inoculated plots were, however, significantly lower in mound number ( $F=8.843$ ) and population index ( $F=14.235$ ) within the third year. *T. solenopsae* spore recovery occurred by the first sampling, 12 weeks post-treatment, and has continued at varying levels through the latest sampling. Highest numbers of spore positive colonies coincided with the third year difference in population between control and inoculated plots. Though this was a small field trial, significant reduction of imported fire ant populations and ability to sustain area infection over a three year span, demonstrates potential for use of *Thelohania solenopsae* as a biological control agent for suppression of imported fire ants.



## A Simulation Model of Competitive Interactions Among Polygyne Red Imported Fire Ant Colonies for Foraging Space and Food Resources

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A simulation model of polygyne (multiple queens) red imported fire ants, *Solenopsis invicta*, was developed. The foraging component integrated the foraging distributions of ants from several colonies in a population with predictions of colony level numerical dominance on simulated food baits. Foraging parameters included the distance to food resources, colony size (i.e. biomass), and average internidal (between nest) spacing among colonies. The colony with the highest potential of having the largest worker force available at a location was considered to be the numerically dominant colony at that location. Three analyses used independent performance statistics derived from observed data to test model performance.

The first analysis showed that the model did a significantly better job of explaining colony numerical dominance on baits than expected from a random assignment of colonies to be numerically dominant on simulated baits. The second analysis showed that the observed and expected frequency distributions of numerical dominance were significantly different when considering numerical dominance as a function of distance from colonies to baits. Food items within 200 cm could be correctly identified as to which colony would be the numerically dominant colony > 80% of time, and declined as the distance between colonies and baits increased. The third analysis showed that the observed and expected frequency distributions of numerical dominance were significantly different when considering numerical dominance as a function of observed ant abundance on baits. The model performed best when there were greater than 50 observed ants on baits.

The second component of the model, food harvesting, relies on parameters from the foraging component coupled with published data on daily energy requirements for colony maintenance and reproduction. The food-harvesting component uses predictions of the daily energy (joules per day) requirements for two food types and two physiological processes. Carbohydrates are required for colony maintenance and proteins for colony reproduction. The food-harvesting component is presented as a hypothetical scenario that dynamically changes each colonies predicted foraging area based on their food harvesting ability and colony demand for food resources. In the simulation, as colonies harvest food their foraging areas shrink in proportion to how much energy they have acquired in relation to an upper limit defined as their daily maximum energy requirement for reproduction or colony maintenance. Simulations depict changes in colony foraging areas as a result of food harvesting under different patterns of resource distributions. (e.g. regular, random, and clumped).



# Endocrine and Ovarian Changes in Newly Dealate Queens of

## *Solenopsis invicta*

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Department of Entomology  
North Carolina State University  
Raleigh, North Carolina 27695

Studies were conducted on the physiological and hormonal changes following the release of alates from developmentally suppressive pheromones produced by mature queens of the fire ant *Solenopsis invicta* Buren. Winged virgin queens were removed from the pheromonal signal and placed in colony fragments. The time for dealation, degree of ovarian development, and biosynthesis rate and whole body content of juvenile hormone (JH) were measured. The production rate and content of JH were highly correlated. Dealation and the initiation of oviposition corresponded to peak production of JH. JH production rose sharply following separation from the natal nest, peaking after three days. After eight days of isolation, JH production gradually subsided to levels similar to that found in pre-release queens, but began to increase again after 12 days. Mature queens had highly elevated levels of JH relative to recently dealate females, probably reflecting the increased reproductive capability of these older females. The results support the hypothesis that the pheromone released by functional queens inhibits reproduction in virgin alates by suppressing corpora allata activity and the production of JH.



# Distribution Patterns of *Thelohania solenopsae* Spores in Red Imported Fire Ant Mounds in Southern Oklahoma

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## Abstract

*Thelohania solenopsae* (Microsporida: Thelohaniidae), a microsporidian, commonly parasitizes fire ants in Brazil and Argentina (Jouvenaz *et al.*, 1981). Recently, attempts have been made to use *T. solenopsae* as a potential biocontrol agent for Red Imported Fire Ant (RIFA), *Solenopsis invicta* (Hymenoptera: Formicidae). This study primarily focuses on detecting the presence of *T. solenopsae* spores in RIFA at three sites in Oklahoma, using two techniques, viz., Polymerase Chain Reaction (Snowden *et al.* 2001) and modified Trichrome staining (Kokoskin *et al.* 1999). In 1998, there were attempts to introduce *T. solenopsae* at two other sites in Bryan County, but the microsporidian spores never established. Similarly, another release in Carter County in 2000 also failed to establish. However, RIFA samples collected in October 2002 from two of our study sites, at Bryan Co., OK tested positive for this microsporidian, while the samples from the third study site at McCurtain Co. (120mi east of Bryan County sites) tested negative. The source of *T. solenopsae* spores in Oklahoma is unknown. Twelve out of fourteen and 17 out of 19 plots tested were positive for the *T. solenopsae* spores, in the two study sites at Bryan Co., and none of the 14 plots tested in the study site at McCurtain Co., had the spores. This study gains significance as this is the first confirmatory report of presence of this pathogen in RIFA workers, in Oklahoma. The results are promising and also widen the scope to study the distribution patterns of this protozoan in the populations of RIFA in Oklahoma.

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# **Influence of Phorid Flies and Low Humidity on Foraging Strategies of *Solenopsis invicta***

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## **Abstract**

The main objective of this research is to investigate the influence of phorids and low humidity on the foraging strategies of *S. invicta*. It is hypothesized that *S. invicta* foraging efficiency will be reduced under parasitoid pressure and low humidity common in a semi-arid desert environment. *Solenopsis invicta* colonies were collected near College Station, TX and Temple, TX. All colonies were polygyne, having multiple queens. The colonies were placed into an arena where foragers were exposed to phorid flies and low humidity. The colonies were starved for 48 hours then allowed to forage for 72 hours. Unlimited treatments were given a fresh three gram piece of hotdog every 24 hours. Food storage piles were located, dried, and weighed at the end of the experiment. *Solenopsis invicta* retrieved more food when food was unlimited regardless of phorid presence or humidity level. Food storage was similar across all treatments. Food was typically stored in the second level of the arena in low humidity and all phorid treatments. *S. invicta* may have overcome phorid pressure by using avoidance strategies.



# Seasonal Effects of Temperature on Red Imported Fire Ants (Hymenoptera: Formicidae)

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## Introduction

Temperature affects many aspects of red imported fire ant, *Solenopsis invicta* Buren, biology including metabolism, development rate, mating behavior, foraging activity, and colony maintenance. We have observed fire ants dying on disturbed mounds and on bare soil during hot, dry weather. An experiment was conducted to determine the effects of hot and cold conditions on fire ant mortality during different times of the year. Critical Thermal (CT) limits are temperatures at which the locomotor ability of the ant is so reduced that it can no longer escape conditions that would lead to its death. The CT limits, critical thermal maxima (CTMax) and critical thermal minima (CTMin), were determined for red imported fire ants in the laboratory over a 17 month period. Fire ants were collected monthly from a local field and tested within 6 hours of collection in a precision controlled computerized incubator affectionately named "Sputnik" (Fig.1).

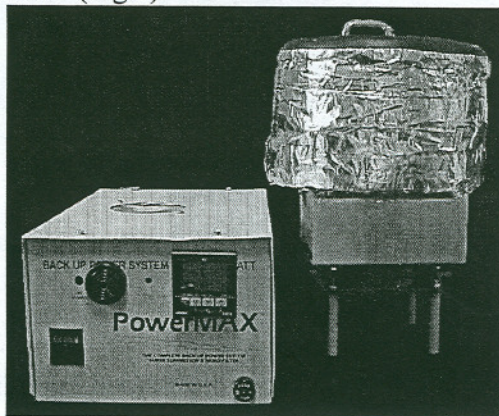


Fig. 1. "Sputnik" – a computerized incubator designed for CT determinations.

## Methods & Materials

Auburn's Research Instrumentation Group designed an apparatus specifically for CT determinations. "Sputnik" is a computer driven thermopile in which an electrical current produces the heat/cold. The computer can be programmed specifically for a task. For example, in our experiment, the computer is programmed to start at a designated temperature (ambient temperature) and raise or lower the temperature 1°C per minute to a destination temperature. A thermister near the bottom of the arena relays constant readings of the arena's interior temperature to the computer while a fan mounted to the Plexiglas® lid circulates air throughout the arena (Figs. 2 and 3).



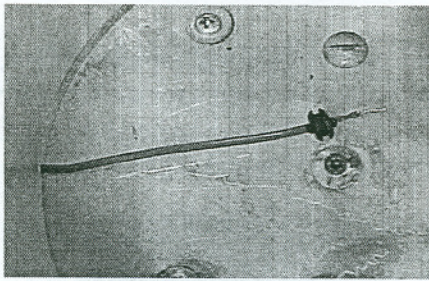


Fig. 2. Thermistor near bottom of arena

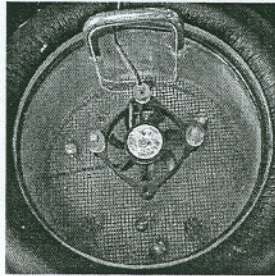


Fig. 3. Fan mounted on Plexiglas® lid

Red imported fire ant workers from monogyne colonies were collected the morning of the experiment and transported to the lab. Workers were individually weighed to the nearest 0.01 mg and placed individually into holding containers. Each holding container consisted of an open-ended glass tube painted with Fluon®. One end of the tube was covered with a piece of no-see-um netting, secured in place by a rubber band (Figs. 4 and 5).

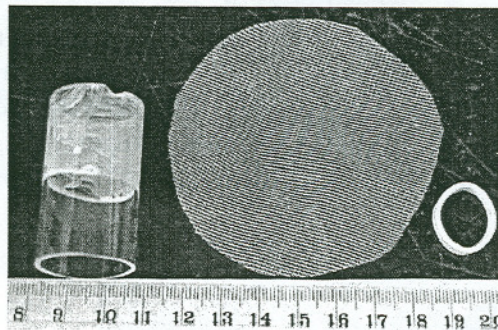


Fig. 4. Fire ant holding container components

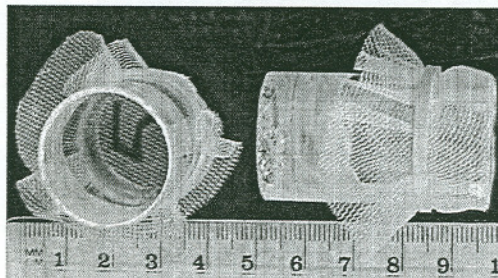


Fig. 5. Assembled fire ant holding containers



Ten numbered holding containers, each containing a single worker, were placed on a raised screen platform in the temperature controlled arena for each run along with a container of water to prevent desiccation (Fig. 6). The arena's temperature is raised or lowered from ambient temperature to determine the CTMax and CTMin, respectively.

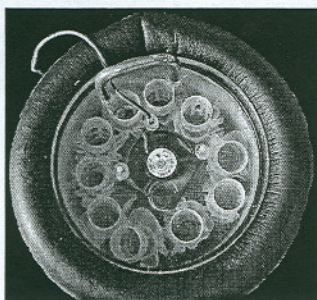


Fig. 6. Loaded arena with lid in place showing holding containers

## Results

The ants were observed until knockdown and the temperature recorded. Knockdown was defined as the temperature at which the ant became inactive or unable to move/right itself (Kay and Whitford 1978; Cockendolpher and Phillips 1990). Once knockdown was determined for a run, the ants were then removed from the arena and placed into a separate holding bin for recovery. 40 CTMax and 40 CTMin for a total of 80 measurements were taken each month for 17 months. Body mass measurements were taken for each ant. Both CTMax and CTMin were plotted against body mass (Figs. 7 and 8). Ant body mass was not related to either CTMax or CTMin at any test period (regression line not shown).

The data indicate that thermal changes in the red imported fire ant are a result of changes in environmental temperatures. Both CTMax and CTMin fluctuated with seasonal temperatures (Figs. 9, 10, and 11). However, a lag time appears to exist between the environment's temperature change and the ants' thermal change. These temperatures are varied, but CTMax's are generally greater than 30°C and CTMin's are 5°C and lower.



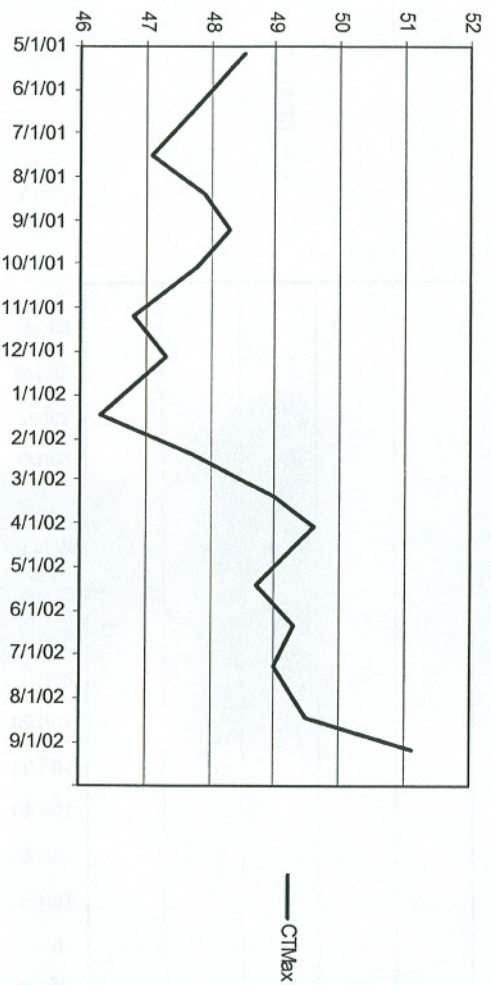
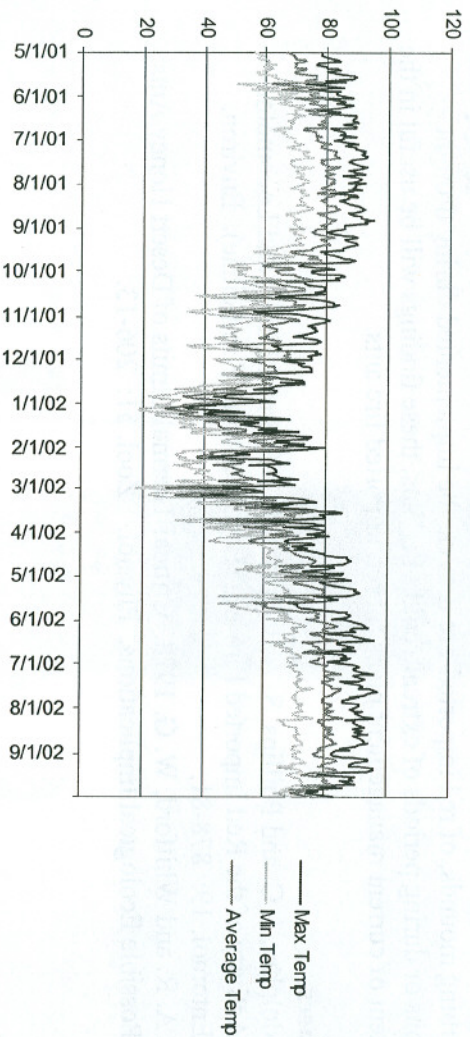


Fig. 9. CTMax temperatures

AWIS Max & Min





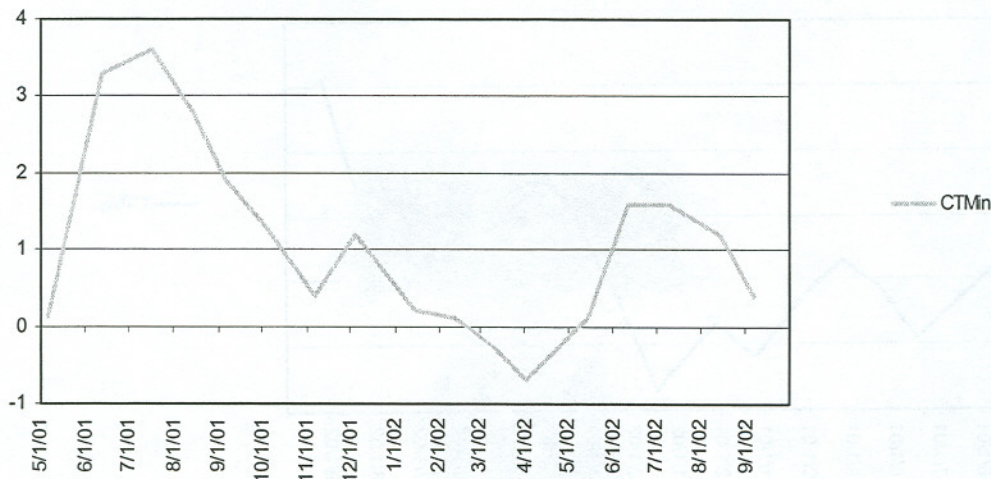


Fig. 11. CTMin temperatures

Ant temperature sensitivity varies with season and could be exploited during rapidly changing environmental conditions. For example, mechanical control, by dragging pastures or disturbing mounds, of red imported fire ants can be implemented during drought conditions or during periods of extreme cold. Perhaps, these findings will be useful in the refinement of current management plans for red imported fire ants.

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# Evaluation of Mechanical Disturbance of Mounds During Cold Weather on Red Imported Fire Ants

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## Introduction

Control of red imported fire ants, *Solenopsis invicta*, using baits or contact insecticides is cost prohibitive in many agricultural settings such as pastures. Shallow disking of a pasture or dragging heavy objects across the pasture are recommended nonchemical management strategies (Drees et. al., 2002). Mechanical control during mild weather conditions usually serves to temporarily flatten hardened mounds and some workers may be damaged. However, disturbing mounds during drought or cold weather may reduce populations by exposing the ants to adverse weather conditions.

The purpose of this study was to evaluate the effect of mechanical disturbance of mounds during cold weather on red imported fire ant populations.

## Materials and Methods

Three treatments were replicated four times in a randomized complete block design. The twelve plots were arranged in a 3.24 ha hay field. Plot size varied within the field to allow for approximately equal numbers of mounds within each plot.

Initial treatments were: 1) {cold} mound disturbance just before a cold front; 2) {warm} mound disturbance during average winter weather; and 3) an undisturbed control. A cold front was defined as a weather system predicted to have several nights below freezing with daily temperatures remaining below 10E C. Average weather was defined as weather with temperatures above 10E C and nights near freezing.

Plots were established and pretreatment population data were collected on October 23, 2001. Mounds were flagged, counted, and rated small (<100), medium (100-10,000) or large (>10,000) based on the number of workers present (Lofgren and Williams, 1982). A garden rake was used to simulate dragging in the cold and warm treatment plots.

Mounds in the warm treatment were disturbed on December 24, 2001 when the daily high was 20E C and the overnight low was -1E C. Mounds in the cold treatment were disturbed on December 26. The daily high was 7.8E C and the overnight low was -5.5E C. Night time temperatures remained at freezing or below until January 11, 2002.

Data were analyzed using the PROC GLM (SAS Institute, 1989b) and means were separated using LSD procedures.



## Results

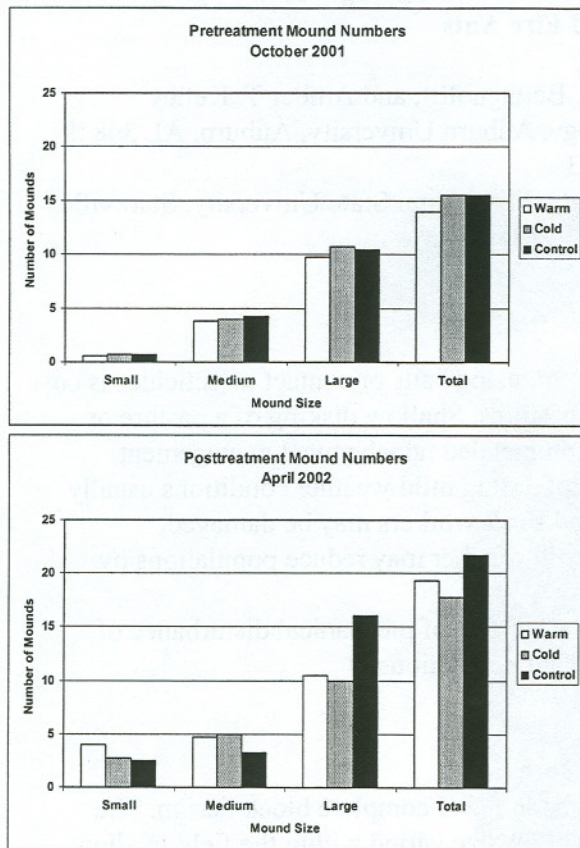


Fig. 1. In all treatments, total mound numbers were higher in April than in October. However, the number of large mounds decreased slightly in the cold treatment and increased in the undisturbed control.



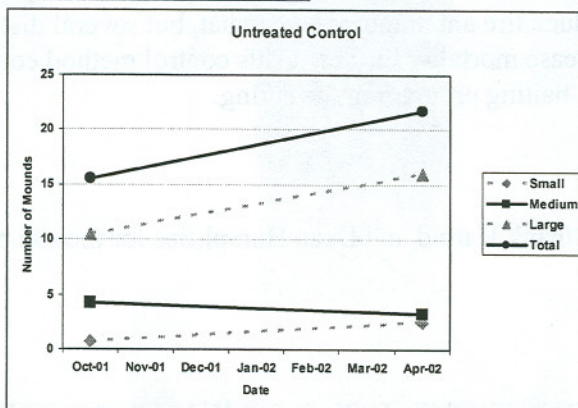
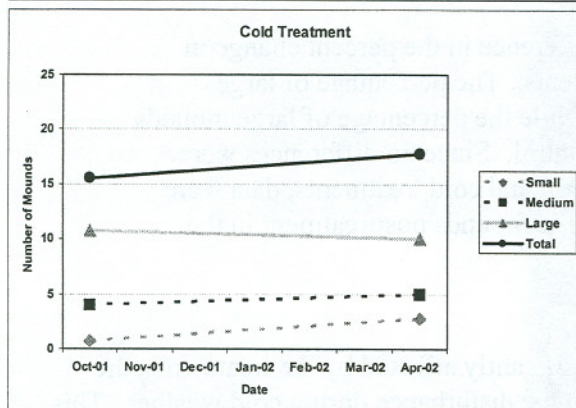
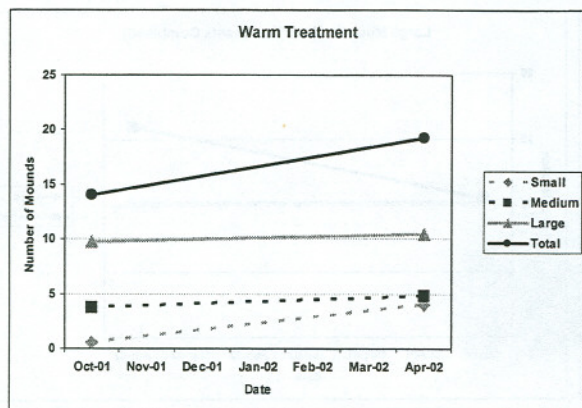


Fig. 2. Numbers of large mounds remained approximately the same pre- and posttreatment in both the warm and cold treatments while numbers of small and medium size mounds tended to increase slightly. This increase in small and medium mounds accounts for the increase in total mounds posttreatment in both treatments. In the untreated control, medium size mound numbers decreased slightly while numbers of large mounds increased, suggesting that the medium size mounds matured during the spring and developed into large mounds.



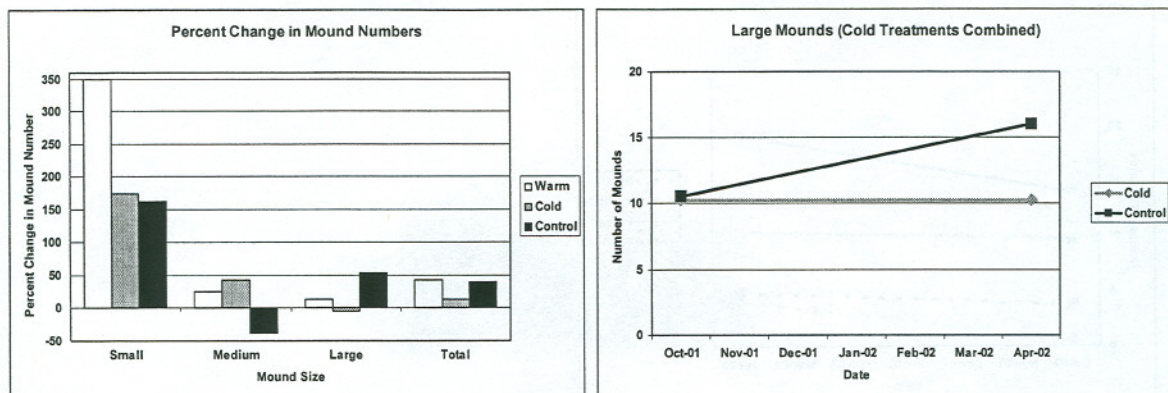


Fig. 3. There was a significant difference in the percent change in numbers of large mounds between treatments. The percentage of large mounds decreased in the cold treatment while the percentage of large mounds increased significantly in the untreated control. Since no differences were found between mound numbers in the warm and cold treatments, data were pooled. There were significantly more large mounds posttreatment in the control than in the disturbed treatments.

While total mound numbers were not significantly affected by the treatments, the number of large mounds decreased in response to the disturbance during cold weather. This method of control will reduce fire ant numbers somewhat, but several disturbances during the winter could possibly increase mortality further. This control method could improve the effectiveness of a planned baiting program in the spring.

## Acknowledgements

The authors would like to thank Harold and Dean Humphries for the use of their land and for help with the project.

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## Red Imported Fire Ants Reduce Lepidopteran Pests in Cotton but not in Soybean

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In previous studies we documented pervasive and often intense effects of predation by the red imported fire ant, *Solenopsis invicta* (Buren) (Hymenoptera: Formicidae), in cotton. Fire ants dramatically decreased the abundance of most arthropod herbivores, including lepidopteran larvae, on cotton plants both in greenhouse experiments and in the field. In this study, we describe greenhouse and field experiments that test for effects of red imported fire ants on lepidopteran larvae in soybean and we contrast these effects with the effects of fire ants in cotton. To quantify the relationship between red imported fire ant abundance and caterpillar abundance in cotton and soybean we sweep-sampled plots in both crops throughout the growing season in 1999. Fire ant abundance had a significant negative effect on lepidopteran larvae in cotton ( $R^2 = 0.27$ ) but not in soybean ( $R^2 = 0.006$ ). Using the field data, we conducted path analyses to quantify the direct and indirect interactions among fire ants, other natural enemies, and lepidopteran larvae. In cotton, the direct effect of fire ants on caterpillars was negative and fairly strong; however, fire ants also negatively affected the abundances of other natural enemies of caterpillars. Despite the indirect positive effects on caterpillar abundance, fire ants still maintained a negative net effect. In contrast, fire ants in soybean had a very weak direct effect on caterpillars, and strong but counteracting direct effects on two natural enemies; therefore fire ants had a very weak net effect on caterpillars. Finally, we conducted greenhouse experiments to quantify the survival of lepidopteran larvae in the presence and absence of red imported fire ants. Significantly fewer caterpillars survived in the presence of fire ants than in the absence of fire ants on cotton plants but there was no difference in caterpillar survival on soybean plants. These results are consistent with the field data that show that fire ants suppress lepidopteran pests in cotton but not in soybean. In cotton, fire ants tend cotton aphids (*Aphis gossypii*) and, as a result, spend more time foraging on foliage, thereby possibly accounting for stronger caterpillar suppression in cotton. Overall fire ant abundance (ground + canopy) was no greater in cotton than in soybean in 1999, but far fewer workers were sampled in soybean foliage. Recently established in the U.S., the soybean aphid (*Aphis glycines*) may provide us with the opportunity to test this idea as it expands its range south into the range of fire ants.



# Non-avoidance of Sodium Bicarbonate-treated Surfaces and Food by the Red Imported Fire Ant

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## INTRODUCTION

Sodium hydrogen carbonate ( $\text{NaHCO}_3$ ), also known as sodium bicarbonate, bicarbonate of soda, or baking soda is a registered pesticide active ingredient for use against fungal phytopathogens (EPA 1999). It controls powdery mildew (Horst et al. 1992) and inhibits growth of yeasts and bacteria (Corral et al. 1988). Effects of  $\text{NaHCO}_3$  on invertebrates are little known, but red imported fire ant, *Solenopsis invicta* Buren, mortality increased with increasing concentration of  $\text{NaHCO}_3$  in laboratory bioassays (Brinkman et al. unpublished data). The objective of this research was to determine whether fire ants are repelled by  $\text{NaHCO}_3$ . If  $\text{NaHCO}_3$  is not repellent to fire ants and causes relatively high levels of mortality, it may be useful as a safe and inexpensive control method.

## METHODS and MATERIALS

Fire ants were obtained from field populations in Spalding County, GA. These ants were removed from soil and maintained as laboratory colonies in plastic trays containing artificial nests. Ants were fed foods including 10% sugar water, tuna in oil, and yellow mealworm, *Tenebrio molitor* L., larvae.

**Arena Choice Tests.** A set of test arenas consisted of two 15.3 cm diam  $\times$  6.5 cm tall plastic dishes connected to a 4.5 cm diam  $\times$  4 cm tall Nalgene container with vinyl tubing. Fluon coated on inside walls of arenas prevented escape of ants. Artificial nests were placed inside each of the large plastic dishes. Sugar water was provided as a food source. Treatments of  $\text{NaHCO}_3$  were evenly spread on the bottom of nests and large containers at a rate of 18.0 mg per  $\text{cm}^2$ . Dental plaster in artificial nests was moistened with water prior to the start of tests; water was added as needed. In one set of arenas, large containers and nests on both sides received  $\text{NaHCO}_3$ . In another set of arenas, neither the containers nor nests were treated with  $\text{NaHCO}_3$ . In remaining arena sets, one side was treated by adding  $\text{NaHCO}_3$  to the large container and nest. All small containers were left untreated. Each test began by adding 100 workers to the small container. Mortality and number of live ants in large containers and artificial nests were recorded each day for 6 d. In the first test, there were five sets of arenas in which only one side was treated with  $\text{NaHCO}_3$  along with one set of arenas in which both sides were untreated (control), and one set of arenas in which both sides were treated with  $\text{NaHCO}_3$ . In subsequent tests, there were two sets of arenas in which either the left or right side was treated with  $\text{NaHCO}_3$ , along with two sets of arenas in which both sides were either untreated (control), or treated with  $\text{NaHCO}_3$ . These tests were conducted a total of three times between 11 June and 21 August 2002 using workers from three different colonies. Experimental design was a randomized complete block design (RCBD). Live ant data for artificial nests and large containers were analyzed separately and were also pooled for comparisons between treated and untreated sides of arena



sets. Data were analyzed by using the PROC MIXED procedure in SAS; significant means were separated with LSD ( $P = 0.05$ ).

**NaHCO<sub>3</sub>-Sugar Water Tests.** Test arenas were prepared by burning a 5-mm diam hole in the bottom of clear 35-ml plastic cups and adding dental plaster to about 10% of total cup volume. Fluon was applied to the inside walls of cups and undersides of lids. Ten workers were placed in each container. Cups were placed on a wet foam pad to maintain moisture in dental plaster within cups. Sugar water treatments were pipetted into 0.65-ml plastic containers. These were placed on the dental plaster in cups. Treatments that were tested were 10% sugar water (untreated control), 10% NaHCO<sub>3</sub> in sugar water, and 10% sugar water in a container placed next to a container with 10% NaHCO<sub>3</sub> in sugar water. Mortality was checked daily for 6 d. Treatments were replicated 10 times in a RCBD. These tests were conducted three times between 18 June and 04 August 2002 using workers from three different colonies. Data were analyzed by using the PROC MIXED procedure in SAS; significant means were separated with LSD ( $P = 0.05$ ).

## RESULTS

**Arena Choice Tests.** Although workers could avoid treated sides, they freely foraged in the large treated containers and also colonized treated nests. Mean number of live workers residing in treated sides (nest + large container) of arena sets did not differ significantly ( $F = 1.12$ ;  $df = 1, 7$ ;  $P = 0.3253$ ) from the number in untreated sides over the 6 days of the test (Fig. 1). The number of workers in both treated and untreated sides declined over time.

On three of the six sampling dates, the number of live ants in treated nests did not differ significantly ( $P > 0.05$ ) from the number in untreated nests. On the other sampling dates, fire ants were present in treated nests, but at lower numbers compared with untreated nests. There may have been some degree of preference for untreated nests, however the reduced presence of ants in treated nests was more likely due to mortality caused by treatments rather than potential repellency of NaHCO<sub>3</sub>. The mean number of workers in untreated nests on day 6 was less than half the number residing in those same nests on day 1.

Mean percent mortality of fire ants in arena sets in which neither side was treated was 25% on day 6 and was significantly ( $F = 22.44$ ;  $df = 2, 5$ ;  $P = 0.0001$ ) lower in comparison to arenas in which one or both sides were treated with NaHCO<sub>3</sub> (Fig 2). There was no significant ( $P > 0.05$ ) difference between mean percent mortality for arena sets in which both sides were treated with NaHCO<sub>3</sub> and mortality in arena sets with only one treated side.

**NaHCO<sub>3</sub>-Sugar Water Tests.** Cumulative mortality for fire ants provided untreated sugar water for 6d was significantly ( $F = 72.96$ ;  $df = 2, 5$ ;  $P = 0.0001$ ) lower than mortality for ants provided NaHCO<sub>3</sub>-sugar water mix and for ants that had a choice between feeding on NaHCO<sub>3</sub>-sugar water mix or untreated sugar water (Fig. 3). Cumulative mortality for fire ants provided sugar water mixed with NaHCO<sub>3</sub> did not differ significantly ( $P > 0.05$ ) from mortality for fire ants that could feed on NaHCO<sub>3</sub>-sugar water mix or untreated sugar water.



## DISCUSSION

The mode of action of  $\text{NaHCO}_3$  in fire ants is unclear. Efficacy of diatomaceous earth against insect pests has been attributed to the action of abrasion and desiccation (Carlson and Ball 1962) and some fire ants may have died from those stressors after foraging in  $\text{NaHCO}_3$ -treated containers. However, Brinkman and Gardner (2001) exposed fire ant workers to diatomaceous earth and observed much lower mortality than was observed for fire ants exposed to  $\text{NaHCO}_3$  in this study. Corral et al. (1988) and Horst et al. (1992) attributed the antimicrobial activity of  $\text{NaHCO}_3$  to bicarbonate ions and elevation in pH. Ants frequently clean appendages and this behavior would have facilitated *per os* entry of  $\text{NaHCO}_3$ . Sodium hydrogen carbonate contributes  $\text{OH}^-$  to solution and large amounts would increase internal pH (Tortora and Grabowski 1996). An increase in internal pH caused by ingestion of  $\text{NaHCO}_3$  may have interfered with enzymatic activity in fire ants and caused their deaths. The optimal pH range for enzymes is narrow (Chapman 1982), and departure from narrow limits of normal  $\text{H}^+$  and  $\text{OH}^-$  concentrations disrupts enzymatic functions (Tortora and Grabowski 1996).

Direct observations of ant feeding were not done, but mortality for ants provided a choice between sugar water and  $\text{NaHCO}_3$ -sugar water mix was higher than for ants provided sugar water only. These results suggest that ants were not repelled by  $\text{NaHCO}_3$  in sugar water and were killed following ingestion. This mortality was relatively moderate (=50%). It is not known if higher levels could be achieved by using higher concentrations of  $\text{NaHCO}_3$  in sugar water or another carrier. If  $\text{NaHCO}_3$  can be distributed by trophallaxis throughout an ant colony, it may be useful as an active ingredient in an ant baiting control strategy.

Sodium hydrogen carbonate or baking soda is safe for humans, inexpensive, and potential exists for use in fire ant IPM. Additional studies should be conducted to determine if  $\text{NaHCO}_3$  can be used in baits and whether it can protect structures such as utility boxes from colonization by fire ants.

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# A SIMULATION MODEL OF COMPETITIVE INTERACTIONS AMONG POLYGYNE FIRE ANT COLONIES FOR FORAGING SPACE AND RESOURCES

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## ABSTRACT

A simulation model of polygyne (multiple queen) fire ant colonies was developed. The foraging component integrated the foraging decisions of ants from several colonies in a population with predictions of colony-level resource distribution on simulated food patches. Foraging parameters included the distance to food resources, colony size (i.e., number), and average intercolony (between nest) spacing among colonies. The colony with the highest potential of having the largest foraging force available at a location was considered to be the numerically dominant colony at that location. These analyses used independent performance statistics derived from observed data to test model predictions.

The first analysis showed that the model did a significantly better job of explaining colony resource distribution on baits than expected from a random assignment of colonies to be numerically dominant on simulated baits. The second analysis showed that the observed and expected frequency distributions of colony size were significantly different when considering numerical dominance as a function of distance from colonies to baits. Third, the model did a better job of explaining colony size than expected from the numerically dominant colony + 80% of time, and distance to the distance between colonies and baits increased. The third analysis showed that the observed and expected frequency distributions of numerical dominance were significantly different when considering numerical dominance as a function of observed ant abundance on baits. The model performed best when there were greater than 50 observed ants on baits. The second component of the model, food harvesting, was an improvement from the foraging component coupled with published data on daily energy requirements for colony maintenance and reproduction. The food-harvesting component uses predictions of the daily energy (calories per day) requirements for two food types and two physiological processes. Carbohydrates are required for colony maintenance and protein for colony reproduction. The food-harvesting component is presented as a hypothetical scenario that asymmetrically changes energy demands predicted foraging areas based on the food harvesting ability and colony demand for food resources. In the simulation, colonies harvested food from foraging areas in proportion to how much energy they have expended in relation to an upper limit defined as the daily maximum energy requirement for reproduction or colony maintenance. Simulation showed that colony foraging areas as a result of food harvesting under different patterns of resource distribution (e.g., regular, random, and clumped).

## INTRODUCTION

Simulation models have been used to investigate many aspects of ant behavior such as nest location (Parker and Goff 1984, Goff and Parker 1984, Impey and Goff 1984, Goff and Parker 1985), chemical recruitment (Dewdney et al. 1990), and information exchange (Vander and Goff 1984). These models describe population level events that are based on the behavior of individual ants. A variety of foraging, recruitment, and nest site models have been developed for ants (Anderson 1980, Rytz and Goff 1980, Goff and Parker 1984, Goff and Parker 1985, Goff 1986, Goff 1987, Goff 1988, Goff 1989, Goff 1990, Goff 1991, Goff 1992, Goff 1993, Goff 1994, Goff 1995, Goff 1996, Goff 1997, Goff 1998, Goff 1999, Goff 2000, Goff 2001, Goff 2002, Goff 2003, Goff 2004, Goff 2005, Goff 2006, Goff 2007, Goff 2008, Goff 2009, Goff 2010, Goff 2011, Goff 2012, Goff 2013, Goff 2014, Goff 2015, Goff 2016, Goff 2017, Goff 2018, Goff 2019, Goff 2020, Goff 2021, Goff 2022, Goff 2023, Goff 2024, Goff 2025, Goff 2026, Goff 2027, Goff 2028, Goff 2029, Goff 2030, Goff 2031, Goff 2032, Goff 2033, Goff 2034, Goff 2035, Goff 2036, Goff 2037, Goff 2038, Goff 2039, Goff 2040, Goff 2041, Goff 2042, Goff 2043, Goff 2044, Goff 2045, Goff 2046, Goff 2047, Goff 2048, Goff 2049, Goff 2050, Goff 2051, Goff 2052, Goff 2053, Goff 2054, Goff 2055, Goff 2056, Goff 2057, Goff 2058, Goff 2059, Goff 2060, Goff 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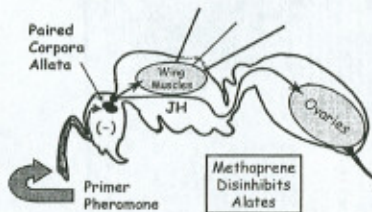
# Endocrine and Ovarian Changes in Newly Dealate Queens of *Solenopsis invicta*

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## INTRODUCTION

Reproduction in fire ant colonies is regulated by the release of chemosignals produced by mature queens. Understanding the properties of these pheromones is integral to developing targeted control measures for this pest species. Recent studies<sup>1,2</sup> indicate that queen primer pheromones may suppress dealation and ovarian activity in alates by inhibiting the production of juvenile hormone (JH) in the corpora allata (CA).

Figure 1. Proposed Model of Primer Pheromone Action



## METHODS

Female *S. invicta* alates collected in Cary, NC were isolated from functional queens for a number of days prior to sampling. The timing of dealation and initial oviposition was noted. Some females were used to determine JH biosynthesis rates by the CA using a radiochemical assay<sup>3</sup>. The CAs were incubated in a growth medium containing tritiated methionine. A time course analysis of JH production indicated an incubation period of 5 hours was optimal. Thin layer chromatography, with *Blattella germanica* samples as a positive control, was used to verify that JHIII was the principle product being measured by the fire ant assay. In addition to having JH biosynthetic rates measured, these females had their ovaries examined for vitellogenic oocytes. The remaining females were homogenized in groups of ten to extract JH for GC/MS analysis<sup>4</sup>, for verification that the rate of JH biosynthesis accurately reflects the hemolymph titer.

## RESULTS

- Fire ant CA produces JH-III
- JH production increases sharply after isolation from natal nest
- Dealation and the initiation of egg production occur subsequent to the rise in JH levels
- High rate of JH production in fecund older queens (sustains increased vitellogenesis?)
- Oocyte production increases steadily for first 15 days, then plateaus

## CONCLUSION

Results suggest that queen primer pheromone suppresses CA activity, thereby preventing dealation and oogenesis. This supports the current model of alate reproductive inhibition.

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Figure 2. Using TLC and the CA's of *B. germanica* as a positive control, the principal product of *S. invicta* CA's is identified as JHIII.

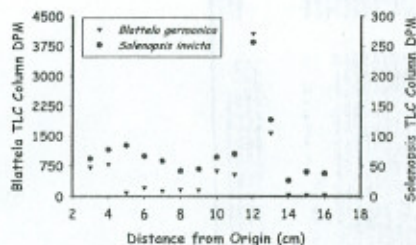


Figure 3. Isolated CA's produce JH linearly at least 6 hrs. CA's in this study are incubated 5 hrs to maximize JH and minimize inter-individual variability.

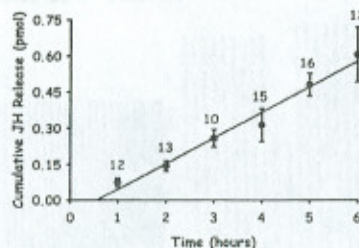


Figure 4. JH biosynthesis rate and hemolymph titer are correlated. Quantities fluctuate after isolation, peaking after 3 and increasing in older females.

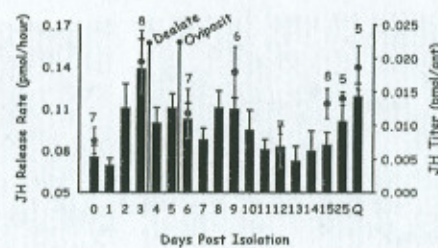
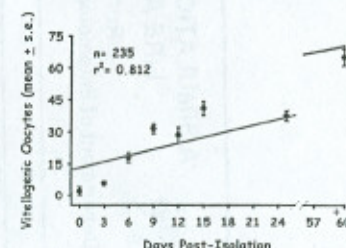


Figure 5. Number of vitellogenic ovarioles increases linearly after isolation. This may be due to rising levels of JH promoting increased rates of vitellogenesis.







# Distribution of *Thelohania solenopsae* in Red Imported Fire Ants in Southern Oklahoma

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## Abstract

*Thelohania solenopsae* (Microsporidia: Thelohanidae), a microsporidium, commonly parasitizes fire ants in Brazil and Argentina (Jouvenaz et al., 1981). Recently, attempts have been made to introduce *T. solenopsae* as a potential biocontrol agent for Red Imported Fire Ant (RIFA), *Solenopsis invicta* (Hymenoptera: Formicidae). This study is a preliminary report on use of two tools, viz., Polymerase Chain Reaction and modified Trichrome staining to detect the presence of *T. solenopsae* spores in RIFA at three sites in Oklahoma. Interestingly, *T. solenopsae* introduced at two other sites in Bryan County in 1998, never established. Similarly, another release in Carter County in 2000 also failed to establish. However, RIFA samples collected in October 2002 from two of our study sites, at Bryan Co., OK tested positive for this microsporidium, while the samples from the third study site at McCurtain Co. (120mi east of Bryan County sites) tested negative. The source of *T. solenopsae* spores in Oklahoma is unknown. Moreover, this is the first instance we have confirmed the presence of this pathogen using PCR and modified Trichrome staining methods. The results are promising and also widens the scope to study the distribution patterns of this protozoan in the populations of RIFA in Oklahoma.

## Introduction

Since its introduction in the 1930s from South America, *S. invicta* has gradually emerged as one of the major invasive insect species in the United States. It infests over 112 million hectares in the United States (Lofgren, 1986) and its impact on the environment is severe. *S. invicta* is found in more abundance in the United States than in its natural habitats in South America (Porter et al., 1992), chiefly due to lack of natural enemies in the introduced habitats. Several workers have recently begun to focus on using sustainable control methods, which mainly includes biological control agents. *T. solenopsae*, a microsporidian obligate intracellular pathogen has been an effective natural enemy of the fire ants in Argentina (Briano et al., 1995). This species has immense potential to control RIFA populations in the United States, also. *T. solenopsae* declines the egg production, queen weight and survivorship of queens and workers of *S. invicta* (Ol and Williams, 2002). In Oklahoma, *S. invicta* is confined to the southern counties, with the exception of few sporadic mounds observed in the northern counties. It will be interesting to track the movement of *S. invicta* populations in Oklahoma in the presence and absence of one of its major natural enemy, *T. solenopsae*. This study is a preliminary report on the distribution of *T. solenopsae* in RIFA populations at three sites in southern Oklahoma.

## Materials and Methods

Fire Ant samples were collected from three sites – two from Bryan Co., OK and the other from McCurtain Co., OK. These sites are ranches where we conduct our Area wide Suppression project studies. Each site has several plots (Table 1) and samples were collected from 3-4 mounds in each plot that had fire ant mounds. The samples were tested for the presence of *Thelohania solenopsae* spores using PCR and modified Trichrome staining.

### Polymerase Chain Reaction:

Fifteen to thirty ants were ground with 500µl TBS using a disposable pestle. To this homogenate, 0.1mm glass beads were added up to three quarters of the tube and beaten at maximum speed for 15 seconds, in a bead beater. The tubes were immediately transferred and kept in 95°C water bath for 5 min. The samples were spun for 10sec at 18000g and the supernatant containing genomic DNA of *T. solenopsae* and *S. invicta* was collected.

2µl of the total DNA was added to the mixture of *T. solenopsae* specific primers - Msp1a and Msp4b (1µl each) and Ready-to-go-beads. This mixture was made up to 25µl with sterile distilled water and the PCR reactions were set following the protocol in Snowden et al., 2002. PCR products were separated on 2% agarose gels and visualized by Ethidium bromide staining (Fig 1). For all experiments, positive (*T. solenopsae* DNA) and negative (PCR reaction mixture without total DNA) controls were also run along with treatments.

### Trichrome staining:

Fifteen to thirty ants were ground with 500µl TBS using a disposable pestle. About 15µl of the ground solution is placed on labeled glass slide and air-dried. The slides are stained using the modified Trichrome staining protocol of Kokoskin et al. (1994). The slides were observed under a light microscope at 400x and 1000x magnifications (Fig 2a&b).

## Results and Discussion

Two of the three sites had *T. solenopsae* spores in the RIFA samples. All the RIFA samples collected from the site in McCurtain county lacked *T. solenopsae*. In Bryan county, this microsporidia was present in 85%-88% of the plots from which RIFA samples were tested. The source for *T. solenopsae* in 2002 RIFA samples in Oklahoma is unknown, as two earlier attempts (1998 and 2000) to establish this pathogen in *S. invicta* mounds failed. The modified Trichrome staining method indicated the presence of this microsporidian spores in RIFA mounds in the plots of Bryan Co., sites. The results from both techniques were in congruence, indicating that *S. invicta* samples can be collected from other counties to study and reliably conclude on the presence (or absence) of *T. solenopsae*.

Table 1. Number of plots sampled and tested for presence of *T. solenopsae* in *S. invicta* mounds at three sites, in Southern Oklahoma.

Study Site	Total number of plots	Number of plots sampled	Number of plots with <i>T. solenopsae</i> *
Adam's Ranch, Bryan County	19	14	12
Bowles' Ranch, Bryan County	22	19	17
McCoy's Ranch, McCurtain County	20	14	0

\* Data are from PCR and modified Trichrome staining methods



Fig 1. PCR products separated in 2% agarose gel. The bands seen in the lanes 1 through 5, 7 and 8 are RIFA samples with *T. solenopsae* spores in Bowles' Ranch, Bryan Co., OK. Lane 6 tested negative for this pathogen. M is the molecular weight marker lane and lanes 9 and 10 are positive and negative controls, respectively.

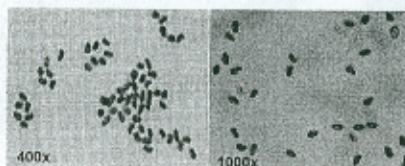


Fig 2a&b. Photomicrograph of binucleate free spores detected from RIFA samples at 400x (2a) and 1000x (2b) magnifications.

## Future Directions

The results from this study are interesting as, the scope for studying the patterns of movement of this pathogen across populations of *S. invicta* is immense. The scenario is very favorable to explore and compare the patterns of movement of *S. invicta* colonies that are infected with *T. solenopsae* and the uninfected colonies. Similar distribution patterns can also be studied for other counties of Oklahoma, where *S. invicta* mounds are prevalent.

## Acknowledgements

VK is thankful to help extended by Dr. Forrest L. Mitchell by providing lab space and supplies to learn PCR and modified Trichrome staining and Jennie Jurney who actually taught these techniques.

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# Eradication Strategies in Orange County, CA - A Neighborhood Plan

Charlie Cassidy, Field Operations Manager and Shana Lowe, Taxonomist / GIS Technician



## Outreach

- Media used to educate BIFA members among citizens
- California Department of Food and Agriculture's (CDFA) create post before is necessary to meet large outdoor in fire and service calls
- Face-to-face interaction with public health confidence
- Notice advise residents of upcoming survey and treatment activities
- OCFAA is well represented at community events



Media support



CDFA create pest hotline



Interface with the community



Property owner notification



Community events

## Survey

- CDFA's eradication surveys outlined 2 major information areas
- OCFAA utilize an average of 5000-6000 chip bucketed baskets weekly
- Bait baskets placed at 50-foot intervals are left at sites for 3-4 hours during optimal foraging periods
- Positive samples identified initiate treatment protocol at site
- Large surveys are conducted efficiently with modified golf carts
- Integrated areas are primary survey sites, but periodic sampling of residential/industrial areas are conducted



Original Infestation Delimited



Sample basket production



Survey flags at 50 ft intervals



Golf cart survey



Wetland survey

## Treatment

- Hydrocortisone is used in a 4-foot area around individual mounds. Pyrethrin fan is broadcast at 1.5 ft/meter
- Sites are treated 4 times at 90-day intervals
- Post treatment survey occurs 90 days after the last treatment
- Post treatment positive sites return to the treatment cycle; negative sites are surveyed semi-annually for 2 years
- "Neighborhood treatment" protocol states that if there are 6 or more positive finds spread within a 1/2 mile buffer, all sites are treated
- All sites within 1/2 mile of positive nurseries are treated
- Many acres/hundreds of homes are treated in one day with use of a modified golf cart/ATV



Half ounce of Dursban® per 1000 sq ft



Treating typical backyard with garden sprayer



Mechanized treatment



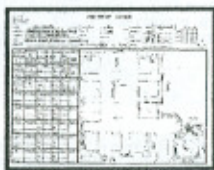
1/2 mile treatment around positive nurseries



Large acreage treated in minutes

## Data Collection

- Original site records were cumbersome and made data retrieval difficult
- Technicians began using Trimble®/Garmin® GPS units in January 2008
- GPS devices are used for data collection and tracking locations
- Data automatically downloads to a computer and can be exported into a database and/or a Geographic Information System (GIS)



Original site record



Electronic data collection in the field



Hand held data collection and GPS unit



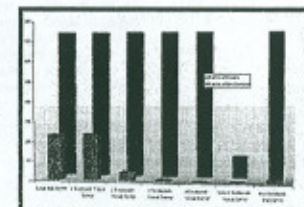
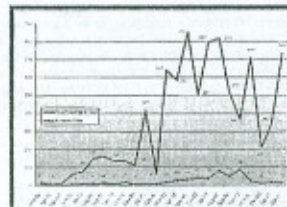
Remote areas can be tracked using GPS



Information downloaded to database/GIS

## Progress

- Countywide post treatment surveys show a 95% reduction in total number of sites after the first treatment cycle
- New colonies are increasingly difficult to locate
- Stinging incidents have become extremely rare





# Influence of Phorid Flies and Low Humidity on Foraging Strategies of *Solenopsis invicta*

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## Introduction

In 1998, *S. invicta* populations were confirmed in southern New Mexico, resulting in a quarantine of Doña Ana County (Plate 1). Although surveys during the past two years have failed to find any *S. invicta* research on potential biological control options are progressing.

Phorids affect their host ants directly, and more importantly, indirectly by influencing worker behavior. Several species of phorid flies are under evaluation, and one, *Pseudacteon tricuspidis*, is being released throughout much of the southeastern US. The influence of low humidity on ant worker behavior and resource retrieval in the presence of parasitoids is still relatively unknown.

## Objective/Hypothesis

The main objective of this research is to investigate the influence of phorids and low humidity on the foraging strategies of *S. invicta*. It is hypothesized that foraging efficiency of *S. invicta* will be reduced under parasitoid pressure and low humidity common in a semi-arid desert environment.



Plate 1. *S. invicta* quarantine map. Doña Ana is the only county quarantined in NM.

## Materials & Methods

*S. invicta* colonies were collected near College Station, TX, and Temple, TX. All colonies were polygynous, having multiple queens. These stock colonies of *S. invicta* were kept at 28°C (RH 85%). Experimental colonies were removed from the stock colonies. Each experimental colony consisted of a fertile queen, 1000 workers, and 10 larvae placed into an 11 liter nesting container. A petri-dish (90mm) with damp lab stone was provided as a nesting site and ants were given water (test tube plugged with cotton). Experimental containers were placed into arenas.

The arenas were designed based on a version of Morrison's (2000) colony level interference competition experiments. The arenas were built with three levels (two enclosed and one exposed to the room environment) (Plate 2).

The lower level was designed to hold containers of water that functioned to maintain high humidity levels similar to an underground colony. The second level supported the experimental colonies in total darkness and was maintained at ~80%RH. The top level had 2 (23x15x5 cm) trays (one buffer tray and one foraging tray) lined with fluen and talc that were connected with 16 cm Tygon tubes (Plate 3). The phorid treatment had a foraging tray (23x15x15cm) covered with saran wrap. The buffer trays were connected to the experimental colonies in the second level using 75 cm Tygon tubes.



Plate 2. Competition arena: lower and second level colonies with 75 cm tubing



Plate 3. Top level: buffer zone (BZ), foraging (F) tray, and phorid foraging (PF) tray

**Resource Retrieval:** Experimental colonies were starved for 48 hours prior to foraging experiments. A three gram piece of hotdog was placed in the center of the foraging tray and colonies were free to forage for 72 hours (Plate 4). Unlimited treatments were given a fresh three gram piece of hotdog every 24 hours. Trays were closed and the number of workers in the foraging tray were counted after 2, 24, 48, and 72 hours of foraging. Five phorids (*Pseudacteon tricuspidis*) were released into the forage tray and new flies added daily if necessary. The hotdog was dried and weighed to estimate foraging. Food storage piles were located, dried, and weighed at the end of the experiment.

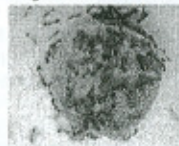


Plate 4. *S. invicta* feeding on top of hotdog



Plate 5. Hotdog covered with fluen by *S. invicta*

## Results / Discussion

The experiment was set up as a split plot design. The food consumption, food storage, and worker movement were analyzed using ANOVA. Means were separated using LSD. Paired columns with different letters are significantly different at P-value < 0.10.

**Foraging:** There were no differences between foraging at high and low humidity. However, when food was limited less food was taken (Figure 1). When food was limited phorids only influenced worker activity at high humidity after 24 hours (Figure 3). At low humidity with unlimited food, phorids significantly reduced worker activity from 24 hours to 72 hours (Figure 4). This suggests that *P. tricuspidis* may have potential in a semi-arid environment. Adult phorid survival was limited to one day even with a food source and was similar between humidity levels. Surprisingly, there was no significance with the extremes, unlimited food at high humidity and limited food at low humidity (Figure 3 & 4).

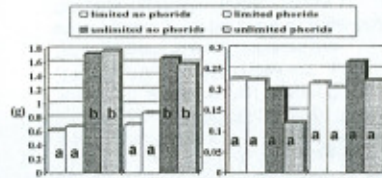


Figure 1. Mean dry weight (g) food retrieved at 80% & 20% RH

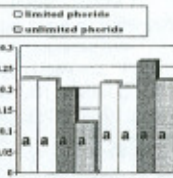


Figure 2. Mean dry weight (g) food stored at 80% & 20% RH

Ants in treatments with no phorid pressure usually fed on top of the hotdog (Plate 4). When colonies were exposed to phorids feeding was usually concentrated underneath the hotdog. Workers may have used this strategy to gather food while limiting risk of being attacked. As noted by others, workers were motionless and had a defensive stance in the presence of phorids. A bizarre behavior occurred in treatments with phorids and also when food was limited. Workers scraped fluen from the trays and piled it on top of the hotdog (Plate 5). Workers may have done this in response to water loss from the hotdog and/or attack from phorids.

**Food Storage:** Approximately 46% of the colonies stored food. Only a small amount of food was stored overall with no significance between treatments (Figure 2). However, with unlimited food, workers stored 12% of total food taken. While on average, workers with limited food stored 31% of total food taken.

In low humidity and all phorid treatments, workers tended to store food closer to the queen. At low humidity with unlimited food this trend was most prominent (Figure 6). At high humidity, regardless of food limitation, workers left a higher percentage of food exposed on the first level of the arena (Figure 5 & 6). Generally, at 24 hours, food was located in the main tube, then slowly moved closer to the queen during the remainder of the experiment.

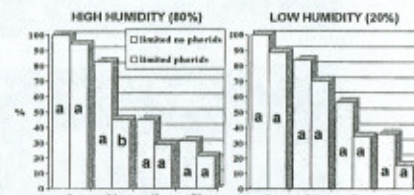


Figure 3. % of workers in forage tray on and associated with food source at 80%RH and 20%RH

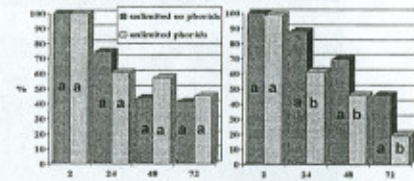


Figure 4. % of workers in forage tray on and associated with food source at 80%RH and 20%RH

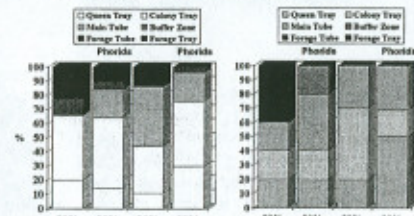


Figure 5. Location % storage for limited no phorids and phorids at 80% & 20% RH

## Conclusions

- S. invicta* retrieved more food when food was unlimited regardless of phorid presence or humidity level.
- Workers were influenced by phorid pressure but had strategies allowing them to still gather food.
- Food storage amount was similar across all treatments. However, workers with limited food stored a greater proportion of food.
- Food was typically stored in the second level of the arena in low humidity and all phorid treatments.

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# Seasonal Effects of Temperature on Red Imported Fire Ants (Hymenoptera: Formicidae)

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## Introduction

Temperature affects many aspects of red imported fire ant, *Solenopsis invicta* Burén, biology including metabolism, development rate, mating behavior, foraging activity, and colony maintenance. We have observed fire ants dying on disturbed mounds and on bare soil during hot, dry weather. An experiment was conducted to determine the effects of hot and cold conditions on fire ant mortality during different times of the year. Critical Thermal (CT) limits are temperatures at which the locomotor ability of the ant is so reduced that it can no longer escape conditions that would lead to its death. The CT limits, critical thermal maxima (CTMax) and critical thermal minima (CTMin), were determined for red imported fire ants in the laboratory over a 17 month period. Fire ants were collected monthly from a local field and tested within 6 hours of collection in a precision controlled computerized incubator affectionately named "Sputnik" (Fig. 1).

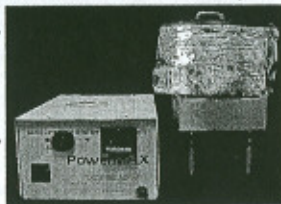


Figure 1. "Sputnik" - a computerized incubator designed for CT determinations.

## Methods & Materials

Auburn's Research Instrumentation Group designed an apparatus specifically for CT determinations. "Sputnik" is a computer driven thermopile in which an electrical current produces the heat/cold. The computer can be programmed specifically for a task. For example, in our experiment, the computer is programmed to start at a designated temperature (ambient temperature) and raise or lower the temperature 1°C per minute to a destination temperature. A thermister near the bottom of the arena relays constant readings of the arena's interior temperature to the computer while a fan mounted to the Plexiglas® lid circulates air throughout the arena (Figs. 2 and 3).



Figure 2. Thermister near bottom of arena.



Figure 3. Fan mounted on Plexiglas® lid.

Red imported fire ant workers from monogyne colonies were collected the morning of the experiment and transported to the lab. Workers were individually weighed to the nearest 0.01 mg and placed individually into holding containers.



Figure 4. Fire ant holding container components.



Figure 5. Assembled fire ant holding containers.

Ten numbered holding containers, each containing a single worker, were placed on a raised screen platform in the temperature controlled arena for each run along with a container of water to prevent desiccation (Fig. 6). The arena's temperature is raised or lowered from ambient temperature to determine the CTMax and CTMin, respectively.



Figure 6. Loaded arena with lid in place showing holding containers.

The ants were observed until knockdown and the temperature recorded. Knockdown was defined as the temperature at which the ant became inactive or unable to move/right itself (Kay and Whitford 1978; Cockendolpher and Phillips 1990). Once knockdown was determined for a run, the ants were then removed from the arena and placed into a separate holding bin for recovery. 40 CTMax and 40 CTMin for a total of 80 measurements were taken each month for 17 months.

## Results

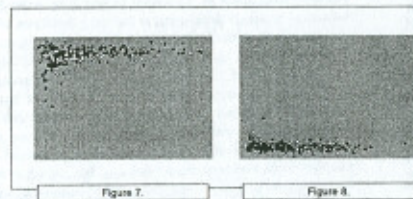


Figure 7.

Figure 8.

Body mass measurements were taken for each ant. Both CTMax and CTMin were plotted against body mass (Figs. 7 and 8). Ant body mass was not related to either CTMax or CTMin at any test period (regression line not shown).

The data indicate that thermal changes in the red imported fire ant are a result of changes in environmental temperatures. Both CTMax and CTMin fluctuated with seasonal temperatures (Figs. 9, 10, and 11). However, a lag time appears to exist between the environment's temperature change and the ants' thermal change. These temperatures are varied, but CTMax's are generally greater than 30°C and CTMin's are 5°C and lower.

Ant temperature sensitivity varies with season and could be exploited during rapidly changing environmental conditions. For example, mechanical control, by dragging pastures or disturbing mounds, of red imported fire ants can be implemented during drought conditions or during periods of extreme cold. Perhaps, these findings will be useful in the refinement of current management plans for red imported fire ants.

## References

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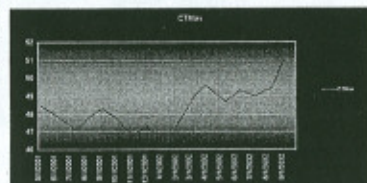


Figure 9.

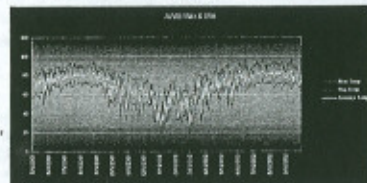


Figure 10.

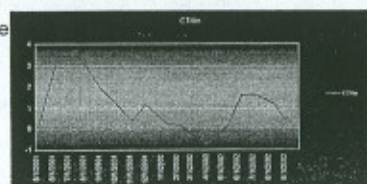


Figure 11.



# Evaluation of Mechanical Disturbance of Mounds During Cold Weather on Red Imported Fire Ants

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## Introduction

Control of red imported fire ants, *Solenopsis invicta*, using baits or contact insecticides is cost prohibitive in many agricultural settings such as pastures. Shallow disking of a pasture or dragging heavy objects across the pasture are recommended nonchemical management strategies (Drees et al., 2002). Mechanical control during mild weather conditions usually serves to temporarily flatten hardened mounds and some workers may be damaged. However, disturbing mounds during drought or cold weather may reduce populations by exposing the ants to adverse weather conditions.

The purpose of this study was to evaluate the effect of mechanical disturbance of mounds during cold weather on red imported fire ant populations.

## Methods and Materials

Three treatments were replicated four times in a randomized complete block design. The twelve plots were arranged in a 3.24 ha hay field. Plot size varied within the field to allow for approximately equal numbers of mounds within each plot.

Initial treatments were: 1) (cold) mound disturbance just before a cold front; 2) (warm) mound disturbance during average winter weather; and 3) an undisturbed control. A cold front was defined as a weather system predicted to have several nights below freezing with daily temperatures remaining below 10°C. Average weather was defined as weather with temperatures above 10°C and nights near freezing.

Plots were established and pretreatment population data were collected on October 23, 2001. Mounds were flagged, counted, and rated small (<100), medium (100-10,000) or large (>10,000) based on the number of workers present (Lofgren and Williams, 1982). A garden rake was used to simulate dragging in the cold and warm treatment plots.

Mounds in the warm treatment were disturbed on December 24, 2001 when the daily high was 20°C and the overnight low was -1°C. Mounds in the cold treatment were disturbed on December 26. The daily high was 7.8°C and the overnight low was -5.5°C. Night time temperatures remained at freezing or below until January 11, 2002.

Data were analyzed using the PROC GLM (SAS Institute, 1989b) and means were separated using LSD procedures.

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SAS Institute Inc. 1989b. SAS/STAT Users Guide, Version 6, Fourth Edition, Volume 2, SAS Institute Inc., Cary, NC.

## Results

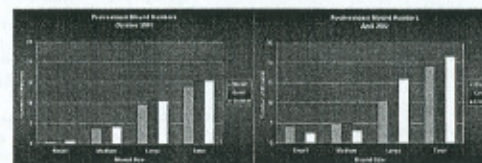


Fig. 1. In all treatments, total mound numbers were higher in April than in October. However, the number of large mounds decreased slightly in the cold treatment and increased in the undisturbed control.

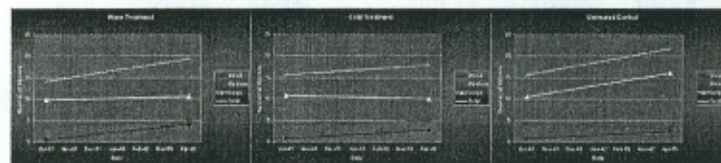


Fig. 2. Numbers of large mounds remained approximately the same pre- and posttreatment in both the warm and cold treatments while numbers of small and medium size mounds tended to increase slightly. This increase in small and medium mounds accounts for the increase in total mounds posttreatment in both treatments. In the untreated control, medium size mound numbers decreased slightly while numbers of large mounds increased, suggesting that the medium size mounds matured during the spring and developed into large mounds.

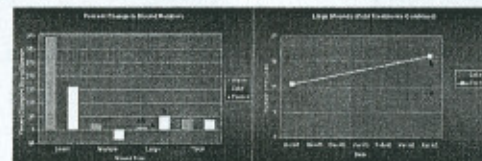


Fig. 3. There was a significant difference in the percent change in numbers of large mounds between treatments. The percentage of large mounds decreased in the cold treatment while the percentage of large mounds increased significantly in the untreated control. Since no differences were found between mound numbers in the warm and cold treatments, data were pooled. There were significantly more mounds posttreatment in the control than in the disturbed treatments.

While total mound numbers were not significantly affected by the treatments, the number of large mounds decreased in response to the disturbance during cold weather. This method of control will reduce fire ant numbers somewhat, but several disturbances during the winter could possibly increase mortality further. This control method could improve the effectiveness of a planned baiting program in the spring.



# Red Imported Fire Ants Reduce Lepidopteran Pests in Cotton But Not in Soybean

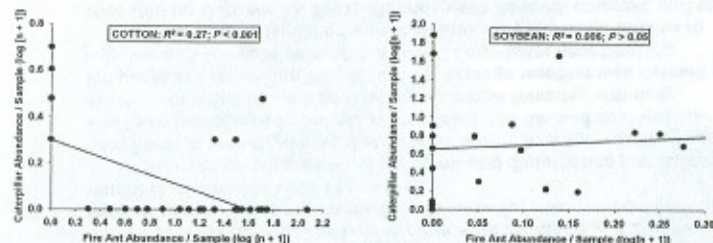
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**INTRODUCTION:** The red imported fire ant (RIFA), *Solenopsis invicta* (Hymenoptera: Formicidae), is an invasive species found in high densities throughout the southeastern United States. *S. invicta* workers are frequently among the most abundant and active predators in agroecosystems, and several studies show that RIFA significantly reduce the abundance of many insect pest species as well as many beneficial arthropods. Cotton and soybean support very similar arthropod food webs but preliminary comparisons suggest differences in the effects of RIFA between the two crop systems. We contrast here the impact of RIFA on lepidopteran pests in cotton and soybean agroecosystems in Alabama.

## OBJECTIVE 1:

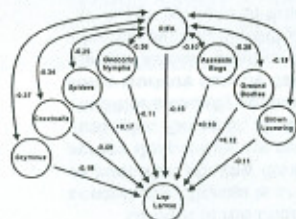
Quantify the relationship between RIFA abundance and caterpillar abundance in cotton and soybean.



Twelve 0.4-ha plots of cotton in Macon Co. and twelve 1.2-ha plots of soybean in Talladega Co. were sweep-sampled for foliage arthropods approximately biweekly throughout the 1999 growing season. Increased RIFA abundance significantly reduced caterpillar abundance in cotton but not in soybean.

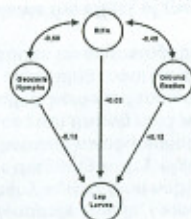
## OBJECTIVE 2:

Quantify the direct and indirect interactions among RIFA, other natural enemies, and lepidopteran pests.



PATH ANALYSIS MODEL IN COTTON

RIFA suppressed the densities of caterpillars as well as the densities of multiple natural enemies that also significantly affected caterpillar density. The net effect of RIFA on lepidopteran larvae in cotton was -0.07.



PATH ANALYSIS MODEL IN SOYBEAN

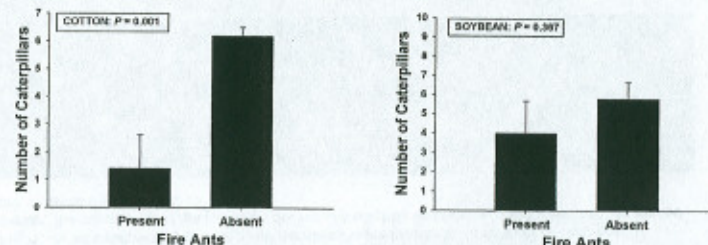
RIFA strongly suppressed the densities of the only two natural enemies that significantly impacted caterpillar densities, but RIFA had practically no direct effect. The net effect of RIFA on lepidopteran larvae in soybean was +0.02.

## OBJECTIVE 3:

Quantify experimentally caterpillar survival on cotton and soybean plants in the presence and absence of RIFA.



In a greenhouse, 12 cotton plants and 10 soybean plants were placed individually in 75 x 30 x 33 cm cages constructed of pvc pipe and mosquito netting. Half of the cages containing either cotton or soybean were connected by plastic tubing to field-collected RIFA colonies to allow workers access to the plants. Seven lepidopteran larvae were placed on each caged cotton plant and 10 larvae were placed on each caged soybean plant. Plants were searched after 24 hours to record the number of caterpillars remaining on the plants.



Significantly fewer caterpillars survived in the presence of RIFA than in the absence of RIFA on cotton plants but there was no difference in caterpillar survival on soybean plants. These results are consistent with the field data (see Objective 1) that show no effect of RIFA on lepidopteran larvae in soybean fields.

## CONCLUSIONS:

1999 field data show that RIFA suppress lepidopteran pests in cotton but not soybean. Path analysis suggests, however, that the direct negative effect of RIFA on lepidopteran pests in cotton is mitigated by RIFA suppression of other natural enemies. In greenhouse experiments, with no other natural enemies present, RIFA significantly reduced caterpillar numbers on cotton plants but not on soybean plants.

In cotton, RIFA tend cotton aphids (*Aphis gossypii*) and, as a result, spend more time foraging on foliage, thereby possibly accounting for stronger cotton pest suppression. Overall RIFA abundance (ground + canopy) was no greater in cotton than in soybean in 1999, but far fewer RIFA were sampled in soybean foliage. Recently established in the U.S., the soybean aphid (*Aphis glycines*) may provide us with the opportunity to test this idea. While we wait for the soybean aphid to invade the Southeast from the Midwest, we are conducting field and greenhouse experiments with artificial honeydew to test whether increased RIFA abundance in soybean foliage suppresses more lepidopteran pests.



# Performance of Organic Insecticides as Individual Mound Treatments Against Red Imported Fire Ants

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Individual mound treatments generally provide the most rapid control of red imported fire ant, *Solenopsis invicta* Buren, colonies. However, mound relocations frequently occur after treatments. Even though many insecticide products are available for fire ant control, most contain synthetic organic insecticides. Homeowners are increasingly concerned about traditional insecticides and therefore are becoming interested in the use of less-toxic or "natural" materials for fire ant control.

Natural insecticides have been used in pest control for centuries. Many are secondary plant substances such as essential oils containing terpenoids. Monoterpenoids such as d-limonene in citrus and l-menthol and menthone in mint add distinctive aromatic characteristics to plants and are used in cosmetics, foods, and as pharmacological additives. Monoterpenoids are toxic and repellent to many insects. In mound drench tests with red imported fire ants, *Solenopsis invicta* Buren, citrus oil formulations containing d-limonene were as effective as a conventional insecticide (Vogt et al. 2002). Abamectin is a mixture of insecticidal compounds derived from natural fermentation products of the soil bacterium *Streptomyces avermitilis*. Abamectin is used to control a variety of insect and mite pests and it is used by homeowners for control of fire ants. Other bacteria also produce natural toxins with insecticidal properties.

The purpose of this study was to evaluate the performance of "natural" compounds against red imported fire ant colonies. Monogyne colonies ( $\approx 12$  inch diameter) located in Lee County, Alabama were treated 24 July 2002 (Tables 1 and 2). Colony (mound) mortality and repellency (relocation and/or satellite mound formation) were monitored 1 day after treatment and at weekly intervals for 28 days. There were 6 replicate mounds per treatment.

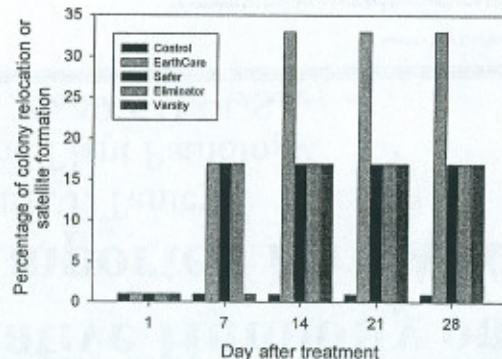
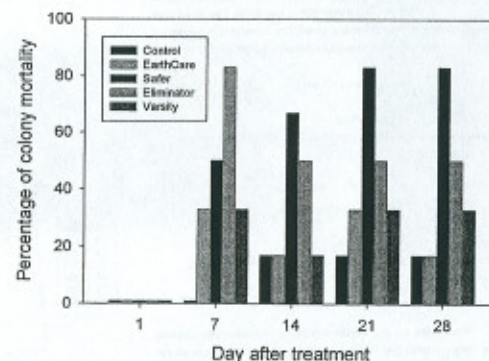
Table 1. Treatments used in this study and their manufacturers and active ingredients.

Treatment	Formulation	Manufacturer	AI	% AI	EPA Reg. No.
Control	---	---	---	---	---
EarthCare	Granule	Spectrum Group	Mint oil	2	---
Safer	Liquid	WoodStream Corp.	d-Limonene	78.2	72244-1-72440
Eliminator	Bait	BioStim	Gram negative bacteria	1	---
Varsity	Bait	Syngenta	Abamectin	0.011	100-893

Table 2. Treatments and application rates and methods.

Treatment	Application Rate	Application Method
Control	---	---
EarthCare	1 cup + 1 gallon water	Granules evenly distributed onto mound then granules watered
Safer	5 oz/gallon water	Dilution evenly sprinkled onto mound; mound drench
Eliminator	1 cup	Bait granules evenly distributed onto mound
Varsity	5 tablespoons	Bait granules evenly distributed onto mound

Eliminator (bacteria) and Safer (d-limonene) were the only formulations that provided  $\geq 50\%$  control of treated mounds at any time period. The Safer formulation provided the best control over the 28 day test. All insecticide formulations were somewhat repellent, however EarthCare (mint oil) granules were the most repellent (33% of colonies relocated or formed satellite mounds). Natural insecticide products applied to individual mounds can provide good control of red imported fire ant colonies. Further research is needed to optimize application rates and methods.





# Effects of Temperature and Relative Humidity on Water Regulation of Alate Red Imported Fire Ants

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The Alabama  
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Management  
Program



**Abstract** Whole-body water loss of alate red imported fire ants was measured in a flow-through system at 10–35°C and 0–100% RH. Water loss increased with increasing temperature and decreasing relative humidity as a paraboloid function of these variables.

## Introduction

During their mating flight and colony founding, alates of the red imported fire ant, *Solenopsis invicta* Burren, are exposed to greater variation in environmental conditions than any other stage. Both temperature and relative humidity (RH) affect net cuticular as well as respiratory water loss. Cuticular permeability in insects generally increases exponentially with temperature (Wigglesworth 1945). Because alate red imported fire ants exhibit a discontinuous gas exchange cycle over a range of temperatures (Vogt and Appel 1990), respiratory water loss can be measured accurately. Respiratory water loss represents 17.5 and 23.0% of total water loss between 15 and 25°C, but only 6.3% loss at 30°C. However, the effects of temperature and RH on total water loss have not been investigated.

The purpose of this study was to quantify the relationship between total water loss in alate red imported fire ants and temperature and RH.

## Materials and Methods

Alate red imported fire ants were collected from monogynous mounds in Auburn, Lee County, Alabama, USA in 1989–1991. Each alate was weighed to the nearest 0.01 mg and placed into a 0.5 ml glass respirometry chamber. Six of these chambers were plumbed in series and positioned within a temperature cabinet maintained at 10, 15, 20, 25, 30, or 35°C. RH of room air was controlled using a Sable Systems (Henderson, NV) DG-1 relative humidity/dew point generator. Air was drawn through the DG-1 relative humidity/dew point generator and into the chambers at 100 ml air<sup>-1</sup> with a mass flow controller and pump (Fig. 1). Alates were re-weighed after a 2 h exposure to a temperature-RH combination. Preliminary experiments indicated that mass loss rates were the same for the first and last alate in a series. Nonlinear regression was used to fit a paraboloid model of the form:

$$z = y_0 + ax + by + cx^2 + dy^2,$$

where  $x$  is temperature in °C,  $y$  is relative humidity in %RH,  $z$  is water loss in mg of water lost per g body mass, and  $a$ ,  $b$ ,  $c$ , and  $d$  are regression coefficients.

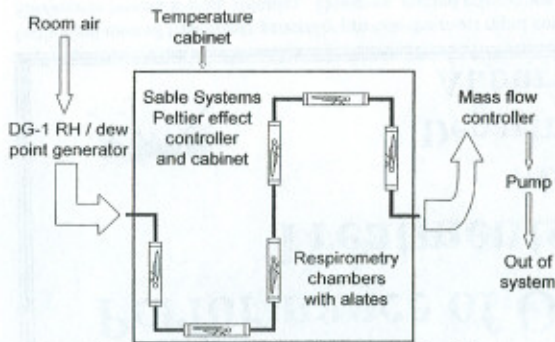


Figure 1. Flow through desiccation system diagram, green arrows indicate air flow.

## Alate Female RIFA

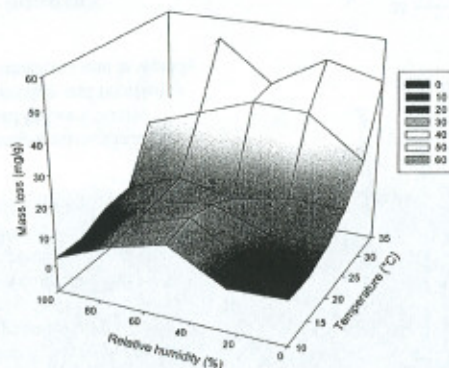


Figure 2. Relationship between water loss of alate female red imported fire ants and desiccation temperature and RH.

## Alate Male RIFA

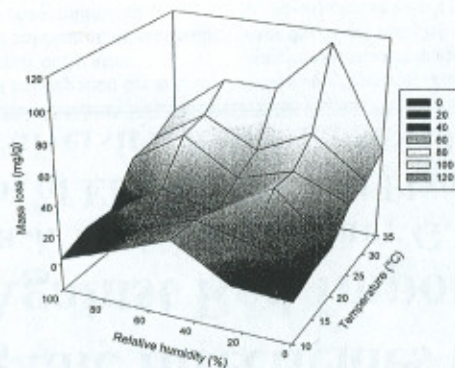


Figure 3. Relationship between water loss of alate male red imported fire ants and desiccation temperature and RH.

## Results and Discussion

Female alate red imported fire ants had significantly ( $P < 0.05$ ) lower 2 h rates of water loss than alate males at virtually every temperature and RH combination. Rates of water loss increased with increasing temperature and decreasing RH for both sexes.

For alate females (Fig. 2):

$$\text{Mass loss (mg/g)} = 23.8 (\pm 7.4) - 2.6 (\pm 0.7) \cdot \text{Temperature} + 0.3 (\pm 0.1) \cdot \text{RH} + 0.09 (\pm 0.02) \cdot \text{Temperature}^2 - 0.003 (\pm 0.001) \cdot \text{RH}^2$$

( $F = 52.91$ ,  $df = 4, 25$ ,  $P = 0.0001$ ,  $r^2 = 0.88$ ).

For alate males (Fig. 3):

$$\text{Mass loss (mg/g)} = 8.0 (\pm 16.4) - 0.9 (\pm 1.6) \cdot \text{Temperature} + 0.9 (\pm 0.2) \cdot \text{RH} + 0.07 (\pm 0.03) \cdot \text{Temperature}^2 - 0.009 (\pm 0.002) \cdot \text{RH}^2$$

( $F = 25.11$ ,  $df = 4, 25$ ,  $P = 0.0001$ ,  $r^2 = 0.77$ ).

Temperature had the greatest influence on alate water loss, nearly 10-fold more than RH. RH tended to affect water loss more at greater temperatures particularly above 25°C. For example, at 35°C female alates lost 46.1 mg/g at 0 %RH, but only 27.1 mg/g at 100 %RH. Whereas at 20°C, female alates lost 8.7 mg/g at 0 %RH and 6.5 mg/g at 100 %.

Increasing temperatures cause both increased cuticular permeabilities and increased rates of respiration. At a constant RH, cuticular permeability and water loss increases curvilinearly with temperature in a variety of insects (e.g., Wigglesworth 1945 and many others). At constant temperature, net water loss increases with decreasing RH, but cuticular permeability remains unchanged (Appel et al. 1986). Respiratory water loss accounts for  $\leq 10\%$  of total loss in alate red imported fire ants between 15 and 25°C and 0 %RH. At 30°C and probably greater, cuticular water loss accounts for  $>93\%$  of total water loss.

These results have important implications for understanding red imported fire ant mating flights, colony founding, and geographical distributions. Temperature is clearly the most important factor in alate water regulation, however, RH can somewhat moderate water loss at higher temperatures.

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# APPARENT FACILITATION OF GROUND-DWELLING MEALYBUGS BY THE RED IMPORTED FIRE ANT, *SOLENOOPSIS INVICTA*

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## INTRODUCTION

Many studies have noted that the red imported fire ant, *Solenopsis invicta*, collects Homoptera honeydew, and it is likely that honeydew is of substantial importance in the diet of this ant (Tennant and Porter 1991, Helms and Vinson 2002). Observations on the nature of interactions between *S. invicta* and Homoptera have been largely anecdotal; however, studies have shown that *S. invicta* will move Homoptera between plants and provide them with protection from natural enemies in agricultural crops (e.g. Vinson and Scarborough 1991, Kaplan and Eubanks 2002). It was proposed recently that ground-dwelling Homoptera, and in particular, an invasive species, the Rhodes-grass mealybug (*Antonina graminis*), may provide *S. invicta* with a significant proportion of necessary carbohydrates in regions of Texas and the southeastern United States (Helms and Vinson 2002). Whether *S. invicta* are simply exploiting these Homoptera for honeydew or whether they, in turn, facilitate abundance of the Homoptera has not previously been studied. As an initial examination, we tested whether the abundance of *Antonina graminis*, and an ecologically similar native mealybug, *Antoninoides* sp., is associated with their proximity to *S. invicta* colonies.

## METHODS

The study was conducted at two sites in eastern Texas during a three-week period in November 2001. One site was located in Brazos Co. and the other in Washington Co. We determined the presence or absence of either *Antonina graminis* or *Antoninoides* sp. on grasses at 5 cm intervals from the center of 23 *S. invicta* mounds to a distance of 3 m from the mounds. The direction of transects from mounds was selected such that the grass species of interest occurred along the entire length and did not pass within 3 m of an adjacent IFA mound. At each 5 cm interval along transects, we selected the nearest root node of bermudagrass (*Cynodon dactylon*), or the nearest plant of oldfield threeawn (*Aristida oligantha*), unearthed the node or plant, and examined the basal area just above the roots for Homoptera. Eleven mounds were using for bermudagrass and 12 for oldfield threeawn.

## RESULTS AND DISCUSSION

*Antonina graminis* occurred on both bermudagrass and oldfield threeawn, while *Antoninoides* sp. occurred only on the threeawn. In both grasses, the presence of mealybugs was associated significantly with distance from *S. invicta* mounds; mealybugs were most common on grasses in mounds and their frequency of occurrence decreased significantly with increasing distance from mounds (Figure 1).

Our results are consistent with the facilitation of ground-dwelling Homoptera by *S. invicta*. The precise nature of that facilitation is unclear; however, there are a number of possible explanations. *Solenopsis invicta* may protect the mealybugs from predators and parasites, and honeydew removal may aid in disease prevention. The fire ant might also actively transport mealybugs during the first instar (crawler) stage. In the study of Helms and Vinson (2002), Homoptera often occurred in shelters constructed by *S. invicta*; however, in this study, shelters were relatively rare. In fact, of all plants with mealybugs, less than 10% had shelters. Why the frequency of shelters varies is unknown; however, it seems clear that shelter absence does not indicate that significant interactions are not occurring (Figure 1).

The relationship between *S. invicta* and *A. graminis* is particularly interesting because both are important invasive species. In the 1940's *A. graminis* was a major pest of rangeland and turf grasses in south Texas. However, a parasitic wasp, *Neodiusmetia sanguani*, was introduced for biological control, and the mealybug was considered "completely controlled" by the 1970's (Dean et al. 1979). While significant economic damage from the mealybug is now apparently infrequent, we know that it is currently widespread and often abundant in the southeast (Helms and Vinson 2000, 2002). It is quite possible that their current abundance is directly linked to association with *S. invicta*.



The Rhodesgrass mealybug, *Antonina graminis*



*Solenopsis invicta* workers tending *Antonina graminis*

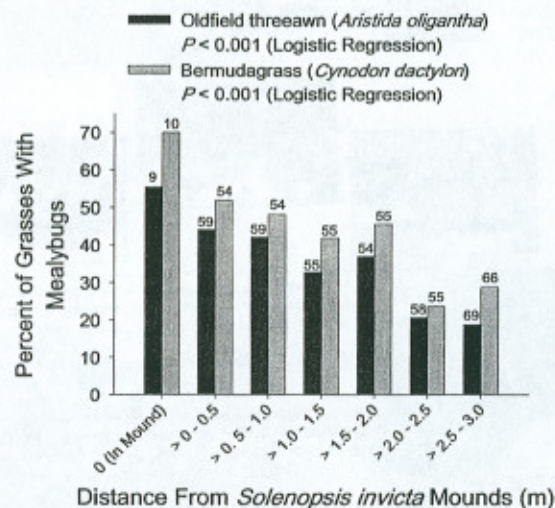


Figure 1. The occurrence of ground-dwelling Homoptera (*Antonina graminis* and *Antoninoides* sp.) on grasses decreases with increasing plant distance from *Solenopsis invicta* colonies. Numbers above the bars are the number of plants assessed within each category. Statistical analyses were conducted on the data at 5 cm intervals.

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# Community Wide Fire Ant Management Programs: Lago Santa Fe

Galveston County, Texas

Cooperators: Lydia L. Heard and Jay M. Gilbert

Paul Nester, Corrie Bowen and Bart M. Drees, Texas Cooperative Extension



## Benefits from Fire ant Community-Wide Program:

Managing red imported fire ants, *Solenopsis invicta* Buren (Hymenoptera: Formicidae) in a larger-area as described by Drees et al. provides more efficient, longer-lasting control (Hooper-Bui et al. 2000). Community-wide management provides several benefits (Riggs et al. 2002):

- Populations were dramatically reduced (91%)
- Control was maintained with repeated applications
- Native competitor ants were maintained or enhanced (6.3 to 9.5 species collected)
- Pesticide costs were reduced for residents (\$35.82 to \$5.86 for bait, 84.5%)
- Fire ant problems were reduced
- Residents' level of knowledge about fire ants increased

## Literature cited:

Drees, B. M., C. L. Barr, S. B. Vinson, R. E. Gold, M. E. Merchant, N. Riggs, L. Lennon, S. Russell and P. Nester. D. Koutoun, B. Sparks, D. Pulett, D. Shedd, K. Vail, K. Flinders, P. M. Horton, D. C. P. L. Keshlar, J. T. Vogt. 2000, revised 2002. Managing imported fire ants in urban areas. B-6043. Texas A&M University, College Station, Texas, 20 pp.  
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Riggs, N. L., L. Lennon, C. L. Barr, B. M. Drees, S. Currenllings, and C. Lent. 2002. Community-wide red imported fire ant programs in Texas (B. M. Drees, ed.), Southwestern Entomologist Supplement No. 25:31-42.



## Demonstrating New Technology:

- Community-wide imported fire ant management using the "Two-Step Method": Step 1) timed broadcast application of an effective bait formulation followed, as necessary by Step 2) use of individual mound treatments (i.e., Orthene®)
- Timed fire ant management beginning April 18, 2003 to achieve maximum control for an athletic event (National Ski Championship, August 13 - 18, 2003)
- Fire ant control around bodies of water sensitive to insecticide run-off
- Application of bait and "Hopper-Blend Treatment": Hydramethylinon fire ant bait (Amdro®Pro or ProBait™) plus methoprene fire ant bait (Extinguish™) at 0.75 lbs of each product per acre
- Application methods: 1) Prototype industrial truck-mounted ant bait "blower" applies 1.5 lbs blended product per acre per minute at 10 miles per hour; 2) GT-77 Herd Seeder mounted on a 4-wheel utility vehicle; 3) Hand-held spreaders to apply baits in fenced areas, ornamental beds and other areas not reachable using vehicle-mounted spreaders



## Documenting Success:

Ant mound numbers per ¼ acre circular plots were used to assess control. Surveys of residents were conducted and information was provided to participants of the event:

- Pre-treatment (4/18): 42.3 mounds/0.25 acre (n = 10)
- 6-weeks: 13.8 (67.5% control)
- 12 weeks: 4.6 (89.1% control)
- July 17: Last assessment
- Aug. 13-18: 2002 National Water Ski Championships!



The Lago Santa Fe Community and the Texas Cooperative Extension would like to thank, Wellmark International, BASF Corporation, Dow AgroSciences, and Ortho for their generous donations of product used in this project.



**RED IMPORTED FIRE ANT CONFERENCE – MARCH 30 – APRIL 1, 2003****PALM SPRINGS, CALIFORNIA****List of Attendees / Registrants****Laura Allen**

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# RED IMPORTED FIRE ANT CONFERENCE – MARCH 30 – APRIL 1, 2003

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**Annual  
Red Imported  
Fire Ant  
Conference**

Palm Springs, CA  
March 30 - April 1, 2003



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**SPECIAL CONTRIBUTORS:**



**Organic Resources, Inc.**

**CALIFORNIA ASSOCIATION OF  
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**Nursery Growers Association**

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**Orange County Vector Control**



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